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Implementation of the Hazard Analysis Critical Control Points (HACCP) Method to Improve Microbiological Food Safety in Peri-Urban Mali

By Ousmane TOURE



Supervisor: Prof. Sandy Cairncross

Disease Control and Vector Biology Unit
Department of Infectious & Tropical Diseases

**London School of Hygiene and Tropical Medicine, Keppel Street, London
WC1E 7HT**

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University of London**

I, the undersigned Ousmane Touré, certify that the research reported in this thesis is my own, except where otherwise indicated.



Abstract

Diarrhoeal diseases remain a main cause of preventable death, particularly among children under five years of age in developing countries. In addition, many studies related to infant diarrhoea causation have demonstrated that the level of contamination is higher in weaning foods than in drinking water. Furthermore, many studies addressed food microbiological contamination and its role in diarrhoea causation. But few of them resulted in an intervention.

Although the Hazard Analysis, Critical Control Point (HACCP) approach has been developed and widely applied to food promotion in industrialised countries, and adapted to small and/or less Developed Businesses, few studies have examined its relevance to domestic preparation of food. However, these latter predicted that the implementation of the approach could lead to an improvement of household bacteriological food safety, but none of them completed the approach to find out how effective it is.

Therefore, this study aimed to take that work one step further, and carried out a small-scale intervention developed on the basis of the HACCP approach. This latter has been extended to health district level in order to find out its impact on microbial reduction in weaning food.

Experiment: The HACCP approach has been applied step by step, to two selected weaning foods prepared by 15 volunteer mothers in peri urban Mali. After setting Critical Control Point (CCP), actions were taken to control, reduce or eliminate microbial growth at these points. 432 food samples were collected and analysed in local Laboratory for FC count to assess the effectiveness of the approach. Lessons learnt were translated into messages delivered in a pilot study.

Pilot study: Sample of 60 volunteer mothers selected randomly was split into two groups of 30, the first undergoing messages directed to actions implementation, and the second standing as a control. Bacteriological samples were taken and analysed and physical parameters were measured, as in the experiment, in 60 households before the intervention and data collected set as baseline.

After three weeks training, alongside with observations, foods samples were taken in both intervention and control households for FC count in local Laboratory.

Flow diagrams of foods, Moni and Fish Soup indicated that they were exposed to contamination at all steps of their preparation and handling. The hazard analysis confirmed FC contamination of all suspected steps except cooking. Four CCPs were identified for each food (cooking, reheating, child service with cooled food after cooking, and child service with cooled food after reheating).

The experiment showed that traditional cooking was very effective in FC elimination; reheating was as effective as cooking when adopted, because no difference existed between two operations' temperatures ($P < 0.0001$). Behavioural corrective actions were effective in controlling FC contamination at remaining CCPs (child service after cooking and child service after reheating).

In conclusion, the HACCP experiment improved significantly the bacterial safety of the two type of weaning foods studied. Thus its behavioural corrective actions were translated into educational messages for the following phase aiming to confirm the effectiveness of the HACCP approach in improving foods safety at household level.

The pilot study data showed the effectiveness of cooking in FC elimination at CCPs considered. A comparison of seasonal variation of FC contamination levels at CCPs showed that these levels were higher at Moni cooking CCP in December (cold season) ($P < 0.0004$) and in August (rainy season) ($P < 0.0002$), compared to June (dry season). They were also higher at Fish Soup storage CCP in December compared to August ($P < 0.0098$). There was significant difference in FC contamination levels between cooking and storage CCPs, the latter was higher than the former, for both Moni and Fish Soup ($P < 0.0001$).

A comparison of FC contamination levels before and after intervention showed that the intervention was very effective in FC contamination reduction at the two remaining CCPs (service after cooking and service after reheating), ($P < 0.0001$). Indeed, at the end of the intervention, contamination levels were less than 10FC/g in more than 83% of cooled food samples (prior to child service) after cooking and about 96% of cooled food samples (prior to child service) after reheating. An assessment of the intervention mothers' ability to perform actions three months later resulted in a better effect, 83 % to 100% of food samples' FC contamination levels met the standard. The present research findings showed that not only was the HACCP approach effective in improving home food safety but also, it was relevant for food hygiene and safety promotion in low income community.

Two research questions were highlighted: firstly, could food safety improvement achieved through the HACCP approach result in diarrhoea morbidity and mortality reduction among young children? And secondly, is the approach scalable and cost effective?

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LIST OF ACRONYMS

AFNOR- Agence Française de Normalisation

B.cereus- Bacillus cereus

°C- Celsius Degree

CAC -Codex Alimentarius Commission

CCP- Critical Control Point

cfu/g- colony forming unit per gram

C. jejuni- Campylobacter jejuni

CP- Critical Point

CSLS- National Programme to fight against HIV/AIDS

DNS- National Directorate of Health in Mali

DNSI- National Directorate of Statistics in Mali

FC- Faecal Coli form

FC/g- Faecal Coliforms per gram

Max- Maximum

Min- Minimum

min- minute

MS/CPS-Ministry of Health/Health Planning Unit in Mali

HACCP- Hazard Analysis Critical Control Point

N- Number (Total)

NASA-US National Aeronautics and Space Administration

ND- Not detected

NS– not significant

NSF-National Sanitation Foundation

RR- Relative Risk

Sdev- Standard deviation

UNICEF- United Nations Children Fund

WHO- World Health Organisation

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CHAPTER I: Food safety and health: A neglected field

Although significant resources have been mobilised these last years by governments, intergovernmental and international organisations, diarrhoeal diseases remain a main cause of preventable death, particularly among children under five years in developing countries. Indeed, it was estimated that, before 1980, 4.6 million, mostly children died from diarrhoea each year. This figure declined between 1980 and 2000 from 3.3 million per year to 2.6 million per year (Keusch et al, 2006).

Boschi-Pinto (2008) estimated the global burden from diarrhoea of children aged less than 5 years at 1.87 million deaths, approximately 19% of total children deaths. In addition, Africa and South- East Asia regions combined contain 78 % (1.46 million) of all diarrhoea deaths occurring among children in the developing world. Moreover, 73% of those deaths are concentrated in just 15 developing countries.

However, diarrhoeal diseases still affect all groups of the population, rich and poor, old and young, in both developed and developing countries. But, a strong relationship exists between poverty, an unhygienic environment, and the prevalence and the severity of diarrhoea episodes for children under five (Kumud et al, 1991; Madhu et al, 1996; Curtis et al, 2000; Keusch et al, 2006).

1.1 Food safety and health in developed countries

In 1996, figures indicated that, infectious diseases accounted for 4% of deaths in the developed world. In the USA alone it was estimated that, each year food borne diseases caused 76 million episodes of illness, 325 000 hospitalisations and 5000 deaths (Mead et al, 1999). In the UK, 2 366 000 cases, 21 138 hospitalisations and 718 deaths, each year are attributed to food borne diseases (Adak et al, 2005). Therefore, despite much progress, infectious diseases remain a big concern in developed countries, (Bloomfield, 2002). Thus, in these Countries, many studies have been conducted to fully understand the routes of contamination and consumers' behaviour, in order to formulate and implement interventions to control infectious diseases, including food borne infections, (Redmond et al, 2003, 2004, 2005, 2006a, 2006b; Worsfold et al, 1994, 1996).

In two studies in the UK, Redmond (2003, 2004) argued that the major risks result from cross contamination behaviours mainly during food preparation, and suggested that quantified risk data could be a valuable tool to prioritise food safety messages. Investigating factors underlying consumers' implementation of specific food safety practices, the same authors (2003) found that there were disparities between participants' knowledge of specific hygiene practices and their implementation of these practices.

In several studies, Cogan (1999), Bloomfield (2002), and Baker (2003) have assessed the effectiveness of hygiene, cleaning and disinfection procedures for the prevention of cross contamination in domestic kitchens, and concluded that rinsing is a critical step in achieving hygiene in the kitchen as well as the use of an antimicrobial agent. One of the studies emphasized the need to better understand hygiene procedures which are likely to be effective in the domestic setting, as well as

the educational and motivational processes needed to promote such procedures. In a home hygiene risk approach study, Bloomfield (2003) highlighted the need to ensure that proper risk benefit assessments are made, and concluded that an approach based on the assumption that “hygiene begins at home” could have far reaching consequences.

Redmond (2005) insisted on the need for the development of consumer-oriented food safety education strategies based on the target audience's perceptions. In an assessment of consumer food safety education provided by local authorities in UK, the same author (2006) found that the leaflet was the most common medium used to inform consumers about food hygiene and that hand-washing, cross- contamination and cooking were the most common issues reportedly addressed in hygiene initiatives. In a pilot study to evaluate the effectiveness of a social marketing-based consumer food safety initiative using observation, the same author (2006) argued that the effect of a smaller immediate intervention upon specific practices could be more effective than the effect of an immediate, moderate intervention upon general food safety practices.

In conclusion, consumers' involvement in the prevention of food borne diseases is crucial because they are the final step in food handling. But much more information is still needed to understand these consumers' behaviours, and the social, economic and cultural factors supporting them.

1.2. Food hygiene and health in Developing countries

1.2.1. Diarrhoea and weaning food

Contrary to developed countries, interventions to reduce bacterial contamination of food have not been fully developed in developing countries (Lanata et al, 2003). The paucity of this kind of interventions is linked to the fact that many people believe water to be the most significant route of transmission of agents causing diarrhoeal diseases. Indeed, it was argued that, up to 90% of childhood diarrhoea is related to poor sanitation, lack of access to clean water, and inadequate personal hygiene (Keusch et al, 2006). Until the late 1980s, poor drinking water quality was considered to be the primary source of diarrhoeal diseases. Furthermore, authors advised public authorities to continue to support safe water supply programs in developing countries. In addition (so ran the argument), domestic hygiene promotion should focus on human stools disposal and effective hand-washing after stool contact (Curtis et al, 2000).

However, in 1989, WHO conducted a review of the interventions for the prevention of diarrhoea among young children, including a review of the literature on the promotion of food hygiene (Esrey & Feachem 1989). So poor was the availability of literature at that time that the reviewers could not draw any conclusion more precise than that, “the limited evidence is fully consistent with the possibilities that a substantial proportion of diarrhoea episodes among young children in developing countries are food borne, related to specific deficiencies in food hygiene, including handling, preparation and storage”. That review assumed that most food borne transmission in developing countries likely takes place within the home, and advocated a major

programme of interdisciplinary research to develop and test cost-effective interventions to promote food hygiene. Furthermore, Lanata (2003) in studies of food hygiene and diarrhoeal disease strengthened the postulate saying that "Food, not water, may be the most important route of transmission of diarrhoea in less developing countries".

For up to 70% of episodes of diarrhoea in developing countries are due to preparation of weaning foods under unhygienic conditions. Infections and associated malnutrition are responsible for an important proportion of the 13 million of deaths among infants and children under five each year. The cause of this morbidity and mortality may be pathogens transmitted through food, (Mortajemi et al, 1993, 1994). Indeed the peak incidence of diarrhoea coincides with the weaning age (Lanata et al, 2003). The highest rate is among infants aged 6 to 11 months (Haggerty et al, 1994; Keusch et al, 2006).

Curtis (2000) hypothesized that behaviours preventing stools from contaminating household surroundings (the child's main habitat), could have greater impact, than those practices preventing pathogens from being ingested. However, many studies related to infant diarrhoea causation have demonstrated that the level of contamination is higher in weaning foods than in drinking water (Esrey and Feachem, 1989; Motarjemi et al, 1993; Lanata et al, 2003). Sheth (2000), in a study of HACCP of weaning foods in an urban slum in Baroda, India, found that infant diarrhoea incidence remained high due to contaminated foods whilst drinking water was found to have no coliforms.

In conclusion this finding indicates that foods could play a significant role in diarrhoeal disease causation, but much more studies are needed to fully understand the epidemiology of food borne diseases, mainly the burden of diarrhoea attributable to food contamination.

1.2.2 Studies of microbiological contamination of weaning foods

The frequency of contamination of weaning foods with pathogens is high in developing countries, (Lanata et al, 2003). This contamination is dependent on many factors, (Henry et al, 1990; Imong et al, 1995; Sheth et al, 2000; Ehiri et al, 2001; Sajilata et al, 2002). The most important of these include: limited resources, illiteracy, poor environmental sanitation and individual hygiene, and cultural and social barriers.

Carefully designed educational programs based on a full understanding of the relationship between behaviours related to these factors, and resulting in feasible and acceptable messages about food hygiene, could lead to food safety improvement.

1.2.3. Microbiological contamination of weaning foods in diarrhoea causation: Some assumptions but little research

Many studies assessed microbiological contamination of food and its role in diarrhoea causation; but few of them resulted in an intervention.

In a study of CCP for foods in households whose members had either alleged typhoid fever or diarrhoea, in Dominican Republic, all stored and non-reheated food samples were found containing FC, *B. cereus* and the risks associated with cooked foods which were not promptly eaten appeared to be greater than that associated with water (Machanie et al, 1988; Sheth et al, 2000).

Henry (1990) in his study of bacterial contamination of weaning foods and drinking water in rural Bangladesh found that “wet” foods such as panta bhat, rice and milk were more contaminated than “dry” foods such as muri and bread. The study highlighted the impact of seasonal variations on foods contamination, confirming that rainy season foods were more contaminated than those of the dry season. No difference in contamination was found between foods stored on the floor or hanging from the roof. Also no difference in contamination was found between foods kept covered and uncovered. In addition, weaning foods contributed most of the faecal bacteria ingested compared to drinking water. The study concluded that, fortunately there is recognition of social, cultural and environmental factors shaping infant feeding patterns.

Ghuliani (1995) studied the influence of the contamination of weaning foods and the transmission of *E.coli* in causation of infantile diarrhoea in low income groups in Chandigarh, India and found a significant correlation between food contamination and diarrhoea history. Indeed, high *E.coli* contamination levels were found in households with diarrhoea history (80.9%), and the maximum incidence of diarrhoea was reported during the weaning period. Unhygienic food storage and subsequent inappropriate heat treatment were reported to be the causes of bacteriological contamination of food. Mothers’ poor individual hygiene was also reported to be a contributing factor.

Imong (1995) examined maternal behaviour and socio-economic influences on the bacterial content of infant weaning foods in rural Northern Thailand, and argued that it seemed likely that the poor food and water quality recorded put infants at risk of contracting diarrhoea. The most important practices contributing to food contamination reported were: mothers’ pre-mastication of foods, hand washing without soap, bottle feeding, prolonged storage of leftover and seasonal foods, and bacterial content variations (foods were more contaminated in rainy season compared to the dry season).

Madhu (1996) studied microbial contamination of weaning foods, and found a high prevalence of *E.coli* in foods in middle income groups compared to those of the high income groups indicating that foods in middle income households were more contaminated than those of high income households and concluded that higher income coupled with better mothers education may result in behaviours favourable to good practices in food preparation and child feeding.

Afifi (1998) in a study of weaning foods contamination where *E. coli*, *B. cereus*, *Shigella* and parasites were counted, in an Egyptian village, detected the first two pathogens in 43.7 % and 21.4 % of 270 food samples taken, respectively. The other pathogens were not detected. The study also found a significant association between food contamination by the two pathogens above-mentioned and the presence of

dung and/or refuse in the house, lack of indoor latrines, non-use of latrines by children, weaning food not freshly prepared, uncovered storage of foods, and the presence of a case of diarrhoea in the house.

In a study of bacterial contamination of *vhuswa*, a traditional maize-based weaning food, and stored drinking water in impoverished households in the Venda region of South Africa, 125 food samples and 125 samples of water were taken and analysed. *E. coli*, *Salmonella*, *Shigella* and *C. jejuni* isolation rates from the *vhuswa* samples were 70%, 5%, 5% and 2% respectively, and total *coliforms*, FC and *F.streptococci* isolation (geometric means) in stored tap-water stored in household containers ranged from 490 to 580 fcu/100 ml, 260 to 370 fcu/100 ml, 310 to 580 fcu/100 ml respectively, and in spring water it was 510 fcu/100 ml, 320 fcu/100 ml and 510 fcu/100 ml respectively, (Potgieter et al, 2005).

Kung'u (2009) in a study of bacteria in complementary foods and drinking water in households with children aged 10-15 months in Zanzibar, Tanzania, comparing a traditional cooked porridge and instant soy-rice porridge, found that the former was less contaminated by *Coliform* and *Enterobacteriaceae* than the latter in both prepared and stored (during 4 hours) foods. In addition, *Coliform* and *Enterobacteriaceae* contamination in instant soy-rice porridge immediately prepared was higher than that of the drinking water. Then, the study concluded that food safety concerns must be addressed when improving complementary foods.

Only Monte (1997) designed and implemented five educational messages (hand washing before and after defined events, boiling water for reconstituting of powdered milk, feeding gruel by spoon rather than bottle-feeding, not storing gruels and milks, and all four together) to improve weaning food hygiene practices of families living in poverty and achieved mothers' behaviour change, despite their limited material resources. Indeed all mothers initiated behaviours and 53 to 80 % of them sustained the new behaviours and practised them every time during a one month period.

Table 1.1 below summarizes weaning food FC contamination levels (fcu/g or 100 ml) in selected studies.

Table 1.1: Faecal Coliforms and other microbial indicators in foods

First author and year	Country	FC in weaning foods (mean cfu/g or % of samples positive)	Drinking water contamination (faecal coliform cfu/100 ml)
Michanie S. 1988	Dominican R.	FC were isolated from 8 of 14 food samples, 5 exceeded 10(5)/g	FC<3 except one sample (from clay vessel) > 9
Henry F J 1990	Bengladesh	Fried ¹ rice (March-July): 2.3; stored boiled rice): 20.5 ¹ after 0-4 h, >87 ¹ after 16 h	
Imong S 1995	Thailand	Rice soup:3.01 ¹ -4.64 ¹	100 samples: 79%
Ghuliani M 1995	India	<i>E coli</i> samples (+) : 56%	
Madhu K 1996	India	<i>E coli</i> samples (+) >49% in low income households and >39% in high income h.	
Afifi ZE 1998	Egypt	270 samples: <i>E. coli</i> (43.7%), <i>B. cereus</i> (21.4%)	
Sheth M 2000	India	Rice (overnight ,25-30°C) :200- 930	
Ehiri JE 2001	Nigeria	Jollof rice: 26 after cooking; 550 after 6h storage	Pipeborne=8.74, borehole=2.57, stream=12.34, spring=3.14.
Potgieter N 2005	South Africa	70%	260-370
Kung'u JK 2009	Tanzania	Stored cooked porridge: 2.24 ¹ Stored soy rice porridge: 4.63 ¹	

¹ Geometric mean²Mean/100 ml

1.2.4. Diarrhoea and infant morbidity and mortality in sub-Saharan Africa: Still a public health problem

In sub-Saharan Africa, diarrhoeal diseases are among the leading causes of mortality and morbidity in young children, despite remarkable improvements in mortality due to better case management and the use of oral dehydration therapy (Haggerty et al, 1994).

From a series of detailed studies in Bobo- Dioulasso, Burkina Faso, Curtis and Kanki (1998), designed the new approach to diarrhoea prevention "Hygiene promotion". This approach is divided into three steps: (i) risk practices (target practices), (ii) motivating behavioural change and (iii) communicating hygiene.

Haggerty (1994) studied a methodological approach in a baseline study of diarrhoeal morbidity in weaning-age children in rural Zaire, and then designed and implemented a community-based hygiene education to reduce diarrhoeal disease and morbidity. The diarrhoeal rates were highest in the first year of life corresponding to the weaning period. Thus, the study concluded that the weaning age had an impact on diarrhoea morbidity. The same author in a study of the influence of demographic, socioeconomic and environmental variables on childhood diarrhoea in a rural area of Zaire, argued that the risk of diarrhoea is associated with age, water quality and sanitation, parental education and household size.

But none of them worked on the improvement of the bacteriological quality of weaning foods. Only Mensah (1990) examined the relevance of cereal fermentation for reduction of bacterial contamination of weaning foods in Ghana, and found that gram negative bacilli were more numerous in unfermented foods (maize dough) than in fermented ones, and concluded that fermentation is an effective method to reduce contamination of maize dough weaning foods with gram negative bacilli.

1.2.5. Diarrhoea and infant morbidity and mortality in West Africa and Mali

According to ORANA (1989) in Burkina Faso 1/3 of morbidity and death among children under 4 years of age is due to diarrhoea. In the Gambia 28.8% of children less than 4 years experienced at least one episode of diarrhoea during the 15 last days of the rainy season, and 15 % of them presented one episode during the dry season. In Mauritania, diarrhoea is the first cause of care seeking for children under 4 years. In Niger one quarter of deaths of children under 4 years is due to diarrhoea. In Senegal diarrhoea is responsible for 10% of the morbidity and 21% of mortality among children under 4 years (ORANA). In all these sahelian countries the peak incidence appears at the age 6- 11 months corresponding to the weaning age.

In Senegal a retrospective study of infants and children (93% were under 3 years) admitted to hospital for diarrhoea over a two- years period, showed that the disease was more frequent in the hot season, and with 10% of the global morbidity, diarrhoea is the primary cause of children's death (ICC Bulletin, 1994). However, in Guinea Bissau a three year cohort study of children under 4 in the peri-urban district of

Bandim confirmed that the peak of diarrhoea morbidity occurs in the rainy season, (ICC Bulletin (1994).

Children's health status is a major indicator of a nation's health, particularly for a developing country like Mali where children constitute about 48 % of the total population. The country's infant mortality rate of 96 per 1000 live births and the under five mortality of 191 per 1000 live births, are among the highest in sub Saharan Africa (MS/CPS, April 2007).

Diarrhoea is the third cause of morbidity of children under five years after Malaria and Acute Respiratory Infections and first reported killer of infants (Konate 2007). 13% of children under five experienced one or several episodes of diarrhoea in the two weeks preceding the recent national Demographic and Health Survey (CPS/MS, April 2007). The age-specific prevalence of diarrhoea, 27% for infants aged between 6 and 11 months is highest, compared to other age groups, (CPS/MS, April 2007).

Breastfeeding is common practice in Mali, because 99 % of mothers of infants under 5 months declared doing so currently. However, only 38 % of infants under 5 months are exclusively breastfed as recommended by WHO and the Ministry of Health; the other infants of this age group received, in addition to breast milk, water (53%), other liquids (4%) and even, in 4% of cases, solid meals or baby food (CPS/MS, April 2007).

Evidence also showed that only 30 % of children aged above 6 months, for whom WHO and the Ministry of Health recommended the use of complementary food, were fed correctly (receiving adequate complementary food) according to these bodies' indications (CPS/MS, April 2007).

It was found that 34% of children under five suffered from chronic malnutrition (under nourishment resulting in delay of child growth) and 16 % suffered from severe malnutrition. It was also highlighted that malnutrition was higher among children aged between 11 and 23 months (20% and 27%), corresponding to the period of weaning, when children are exposed to illnesses due to unhygienic weaning food and exposure to an unhealthy environment (CPS/MS, April 2007).

In Mali, 25,542 persons among whom 1,481 are children were recorded suffering from HIV/AIDS (CSLS, June 200). These people due to their immune-depressive status are highly vulnerable to bacteriological infection while food supplementation was one of the important factors for improving their health status (WHO/UNICEF, 2008).

Diarrhoea is still a major public health problem in West Africa in general, and in Mali in particular, and sound and integrated strategies are needed to tackle the problem.

1.3. HACCP and its relevance in food safety

The HACCP (Hazard Analysis, Critical Control Point) approach has been developed and widely applied to food promotion in industrialised countries, (Mitchell, et al 1992; Little et al, 2003, 2004; CAC/RCP, rev4- 2003) and adapted to Small and/or less

Developed Businesses (WHO, 1999, FAO/WHO, 2005). The approach was even applied as a management tool for monitoring and controlling microbiological hazards in water treatment facilities in South Africa; CCPs identified and controlled were: raw source water, sedimentation, filtration and chlorine-disinfection. The experiment resulted in the delivery of water meeting safety standards, however control at certain CCPS (sedimentation and filtration) failed to meet criteria (Jagals et al, 2004); moreover the HACCP approach has been incorporated into the WHO Drinking Water Quality Guidelines. But few studies have examined its relevance to domestic preparation of food.

Griffith (1994) in a study of an assessment of the potential of the application of HACCP to food preparation practices in domestic kitchens concluded that the implementation of the approach in home food preparation could be beneficial to all countries. The same author (1997), in the assessment of the standard of consumer food safety behaviour, argued that the HACCP system could provide a methodological tool and draw attention to preparation steps that were critical for food safety.

Ehiri (2001), in another observational study, applied the approach to the preparation and handling of five weaning foods in eastern Nigeria, and identified three critical control points common to all processed heating foods: cooking, storage and reheating; other critical control points were added for some foods depending on specific complementary processes they underwent before or after cooking, for instance adding ingredients. In all foods examined, microbiological contamination was higher before cooking and during six hours storage (contamination range: 200-600 cfu/g, and 30-1300 cfu/g respectively) than after cooking (range:0-81cfu/g).

Sheth (2000) conducted a study in an urban slum "Kalyannagar" in Baroda, India, using the same approach to five local foods and found critical control points similar to those mentioned by Ehiri: cooking and storage for the chappati food. All these studies predicted that the implementation of the approach could lead to an improvement of household bacteriological food safety, but none of them completed the approach to find out how effective it is. Table 1.2 summarizes selected studies' findings.

In 1992 a FAO/WHO Joint Expert Committee on Food Safety recommended the use of HACCP in household studies in developing countries so that information collected about food-associated hazards could be directed to health education programs, (Bryan, 1992). This study aimed to implement that recommendation and take the work one step further, and carried out a small-scale intervention developed on the basis of the HACCP approach. This latter has been extended to a larger sample selected at a health district level in order to find out its impact on microbial reduction in weaning food.

Table 1.2: CCPs, control measures and monitoring criteria of some foods

First author	Country	Food and its CCPs found	Control measures	Monitoring
Bryan (1992)	Not defined	Rice: cooking, storage, reheating and service	Cook thoroughly, serve promptly, hold >55°C, cool rapidly and reheat thoroughly	Temperature and time
Bryan (1992)	Not defined	Infant formula from milk: boiling water, milk-can opening and storage.	Rinse bottle properly and holding temperature	Temperature and time
Sheth (2000)	India	Chappati: cooking, storage	Not defined	Not defined
Ehiri (2001)	Nigeria	Jollof rice: cooking, storage and reheating	Cook thoroughly, serve as soon as prepared, reheat thoroughly	Temperature, colour, wash hands with soap, wash utensils and cooking facilities
Ehiri (2001)	Nigeria	Moi-moi: cooking, storage and reheating	Prepare at home, cook thoroughly, serve as soon as prepared, reheat thoroughly	Temperature, colour, wash hands with soap, wash utensils and cooking facilities
Ehiri (2001)	Nigeria	Agidi: cooking, storage and reheating	Prepare at home, cook thoroughly, serve as soon as prepared, reheat thoroughly	Temperature, colour, wash hands with soap, wash utensils and cooking facilities
Ehiri (2001)	Nigeria	Soybean powder: purchasing, ready to use, handling and storage	Prepare at home, hands washing with soap, utensils and cooking facilities washing, separate from raw foods, serve as soon as prepared and handling minimally	Washing hands with soap, washing utensils and limit storage
Ehiri (2001)	Nigeria	Ground crayfish: handling and storage	Prepare at home, hands washing with soap, utensils and cooking facilities washing, separate from raw foods, serve as soon as prepared and handling minimally	Temperature, colour, wash hands with soap, wash utensils and cooking facilities, and limit storage

1.4. HACCP History

HACCP was developed in the 1960s, in response to NASA's need to increase confidence that food supplied to astronauts was safe. But it has gained international recognition with industry and governmental bodies as the system of choice for food safety (NSF, 2006). Besides, the Codex Alimentarius Commission (CAC) adopted the HACCP as part of its Recommended International Code of Practice – General Principles of Food Hygiene guidelines (1969 rev4 2003). Since then, it is considered as one of the most relevant tools to ensure food safety and widely used to certify the quality of foods intended for international trade and large scale manufacture.

1.5. The Hazard Analysis Critical Control Point (HACCP) approach

HACCP is a systematic approach to identify, assess and control hazards. It seeks to identify hazards associated with any step of food production, preparation, and handling, assess the related risks and forecast control procedures needed. It is composed of 7 steps, Bryan (1992); however five preliminary steps are needed to meet the conditions of its implementation (NSF, 2006).

The following list and Figure 1.1 below show the logical sequence for the application of HACCP.

1.5.1. Five preliminary steps before HACCP

1.5.1.1. Assemble the HACCP Team

The HACCP team is a group of professionals who understand both HACCP and the domain of the study and decide to conduct a HACCP plan (NSF, 2006). The HACCP team establishes the scope of the HACCP by identifying what product and processes it covers.

1.5.1.2. Describe product

This description includes: Raw materials and ingredients, preparation processes and storage.

1.5.1.3. Identify product intended use

Regarding the expected use of the prepared foods, information needed is mainly: importance of foods in the diet of the target group and handling practices.

1.5.1.4. Construct a flow diagram:

This is a flow chart for each selected food, representing each operation by a rectangle with arrows to indicate direction of flow, Frank (1992). Each step in the process covered by the HACCP plan must be outlined and process and physical location listed (NSF, 2006).

1.5.1.5. On- site confirmation of the flow diagram:

These are the processes to confirm that all steps are identified and accurately described by the flow diagram.

1.5.2. Seven steps of HACCP process itself

1.5.2.1. Conduct a hazard analysis:

This aims to identify the hazards and assess their severity and risks associated with them. It is the process of identifying significant risks relative to the food product or handling processes. It takes into consideration the hazards associated with the intended end use of the food. This step is critical to the success of the HACCP plan because it serves as the basis for the rest of the HACCP activities (NSF, 2006)

1.5.2.2. Identify the Critical Control Points (CCP):

A critical control point (CCP) is an operation at which action (control) must be exercised over one or more factors to eliminate prevent or minimize a hazard, (Bryan, 1992). Figure 1.2 below shows an example of a decision tree to identify CCPs (NSF, 2006).

1.5.2.3. Establish critical limits:

A critical limit is a measurement or observation that separates what is acceptable from what is not acceptable (e.g. $> 60^{\circ}\text{C}$ for at least 12 min). This critical limit cannot be violated if the hazard has to be controlled at that CCP. Critical limits must be effective at keeping the hazard under control. Critical limits can be quantitative (numerical) or qualitative (descriptive).

1.5.2.4. Monitoring:

This involves systematic observation, measurement and/or recording of the significant factors for control of the hazard. Procedures chosen must permit action to be taken before the food is made available to the consumer.

1.5.2.5. Establish corrective actions:

These are actions to be implemented when monitoring indicates that criteria set for safety and quality at a particular critical control point are not met (Bryan, 1992).

1.5.2.6. Verification:

The WHO defines verification as “the application of methods, procedures, tests and other evaluations, in addition to monitoring to determine compliance with the HACCP plan” (NSF, 2006). It encompasses collection of information and tests to ensure that the system is working as planned (Bryan, 1992).

1.5.2.7. Record keeping

The record keeping consists of the compilation of all data related to the scope of the HACCP plan.

Figure 1.1: Logic sequence for the application of HACCP (NSF, 2006)

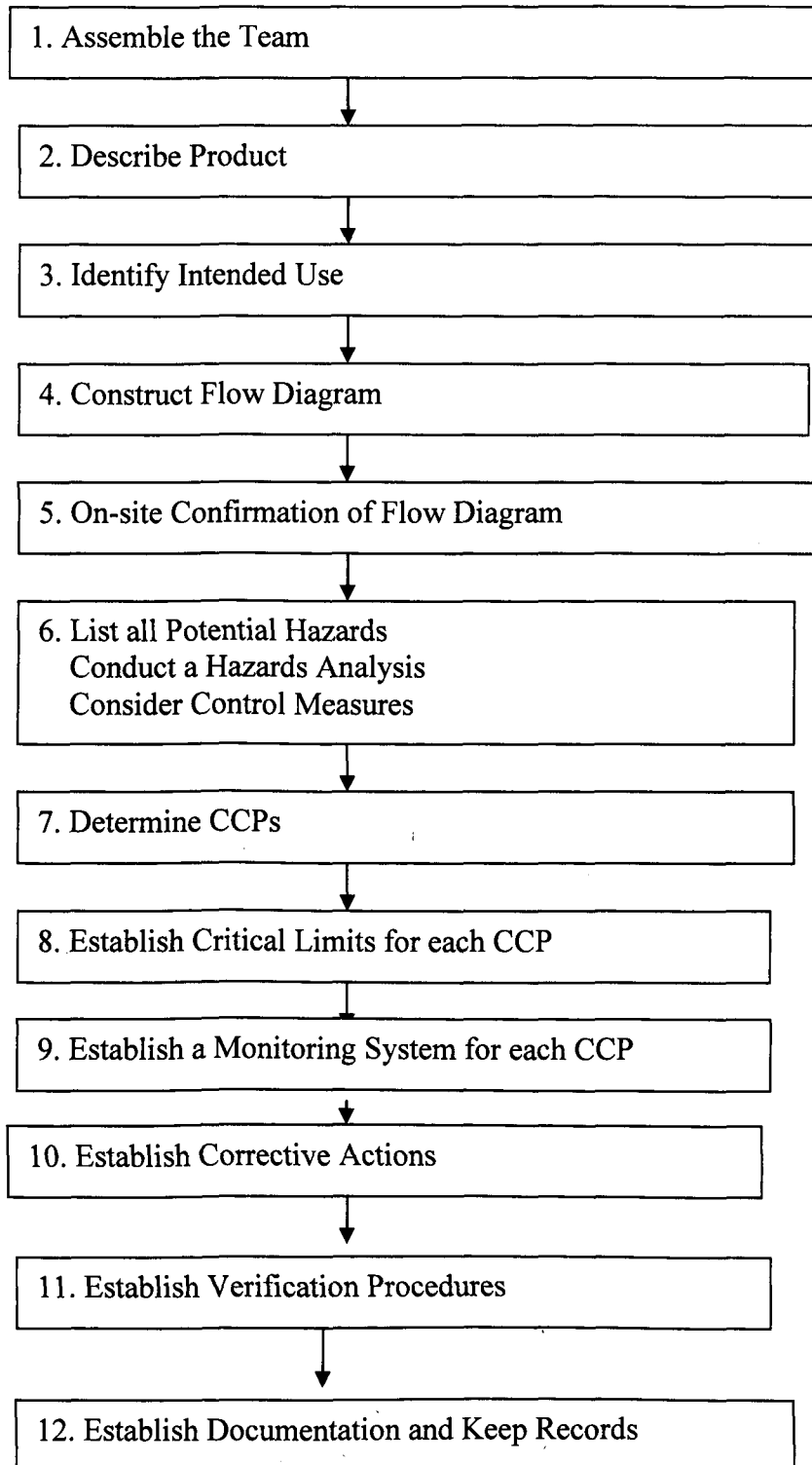
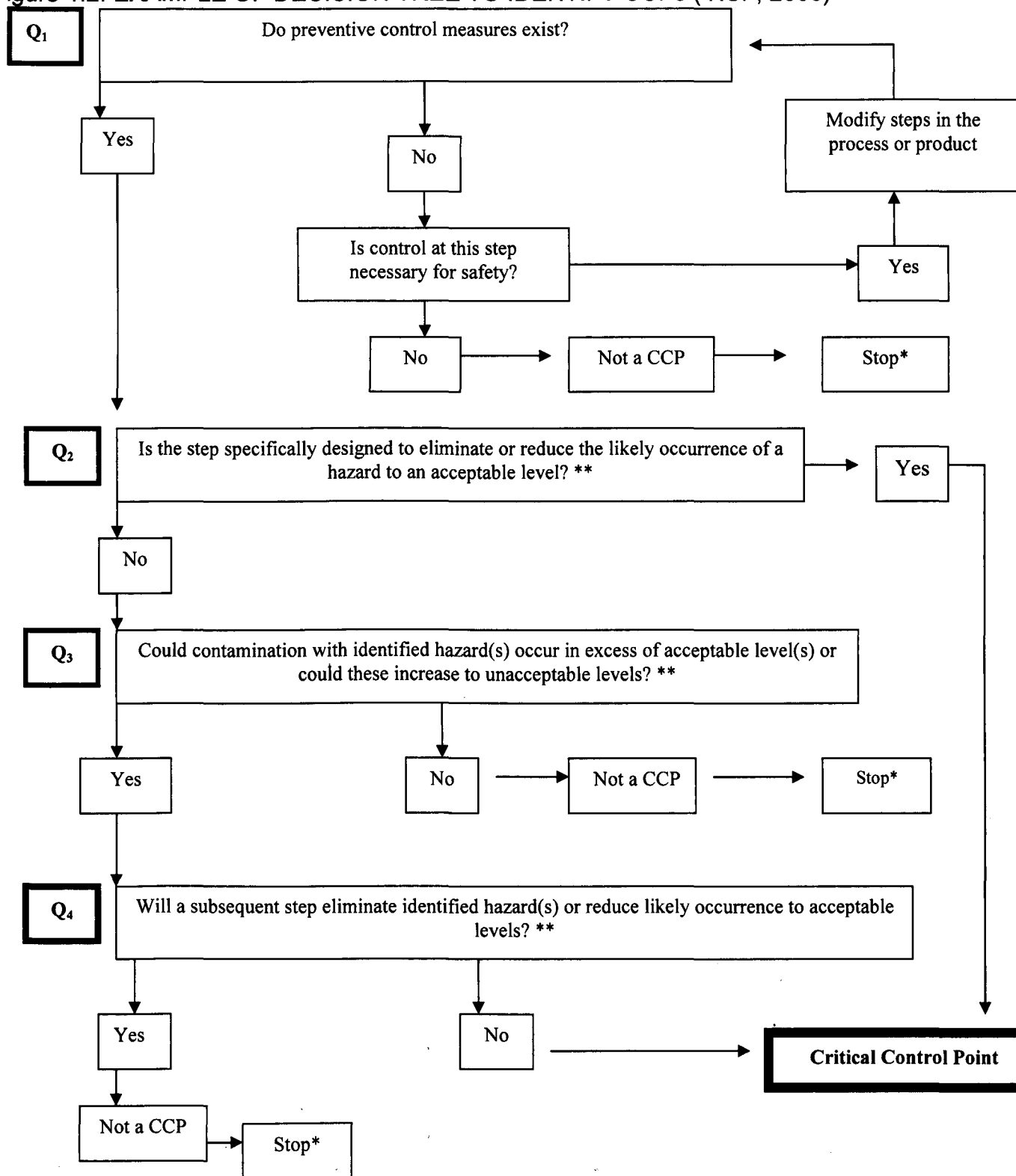


Figure 1.2: EXAMPLE OF DECISION TREE TO IDENTIFY CCPs (NSF, 2006)



* Proceed to the next identified hazard in the described process

** Acceptable and unacceptable levels need to be defined within the objectives in identifying the CCPs of HACCP plans

1.6. Study objectives and Thesis outline

1.6.1. Study rationale, aim and objectives

The study aims to find out the effectiveness of HACCP procedures in improving household food safety by applying the approach to weaning foods in peri-urban Mali in order to draw lessons which can be directed to health education programmes.

The specific objectives are the following:

- Construct the flow diagram of two major foods used as weaning foods ;
- Test small scale HACCP approach in which mothers' food preparation is supervised, to draw lessons on how to improve microbiological safety of the two selected foods ;
- Take the experiment nearer to reality, by observing the mothers' food preparation for compliance with previously delivered guidance.

The project has been developed in two phases; the first phase, the experiment study, targeted the achievement of the first two objectives and the second phase aimed to achieve the last specific objective.

For practical reasons this study has been limited to microbiological hazards only; and even in this case microbial spore formation and growth as well as protozoa pathogens were excluded.

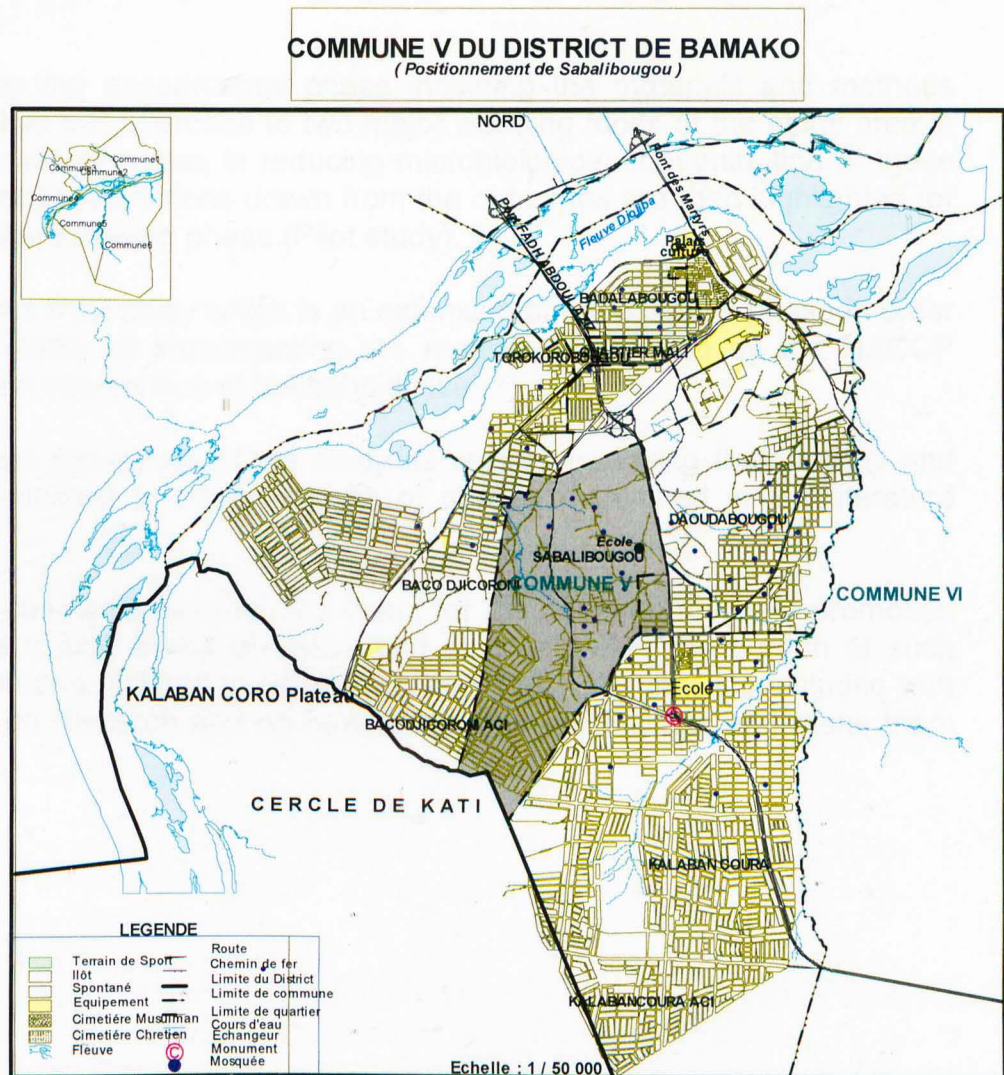
1.6.2. Selection of the site

Mali is a sahelian landlocked sub-Saharan Africa Country located in the West Region. Its capital is Bamako. The latter is crossed from East to West by the Niger River. The city of Bamako is divided into six communes led by elected Mayors. Located on the right side of the River, Commune V, the area of our study is divided itself into nine zones (Bacodjicironi, Sabalibougou, Quartier-mali, Torokorobougou, Bacodjicironi-ACI, Badalabougou, Kalabancoro, Daoudabougou and Garanribougou)

The first phase took place in Sabalibougou, one of the poorest zones with regard to the availability of urban basic services (water supply systems, sanitation, housing...). The zone is hilly and covers 4.5 square kilometres for a population of 50,269 inhabitants. 95% of the population is Muslim. The biggest local market of the commune is located in this zone; the main activity of inhabitants is small informal trade. 75% of traders are female and the majority of males are manual workers. The community water supply reaches a small part of the area; most people buy potable water from itinerant vendors who buy their water at community bore holes (52 bore holes in total), which are managed by rich individuals who generally do not live in the zone.

The second phase of the study related to the whole of Commune V with a total population about 286,723 inhabitants. Socio-economic indicators for the other zones are similar to those described in the experimental zone. The health care system encompasses one reference health centre under the responsibility of the local authority, nine community health centres managed by community health centre's associations (one for each health centre) and 27 private health centres. Malaria, acute respiratory infections and diarrhoea are the major health problems of the area. Only 14.3% of the population get their potable water from the city's water supply system; the rest is provided with water by private wells (20.6%), community boreholes (44.69%) and vendors.

Figure 1.3 : Map of the study area (commune V). Sabalibougou is shown shaded



1.6.3. Thesis outline

This thesis is divided into four chapters:

The first chapter covers a literature review of studies related to food borne diseases and bacteriological contamination of foods in both developed and developing countries, with particular emphasis on diarrhoea among children under 5 in sub-Saharan Africa. It highlights the severity of the problem of contamination of weaning foods of developing countries, its alleged role in diarrhoea causation and the need for more data. The relevance of the HACCP approach to food safety is highlighted to support our use of it. Finally, the aim, rationale and setting of the study are presented.

Chapter 2 presents the experimental phase including the materials and methods used to apply the HACCP approach to two major weaning foods of the study area in order to assess its effectiveness in reducing microbiological contamination of these foods. Relevant corrective actions drawn from the outcomes are then highlighted for implementation in the following phase (Pilot study).

Chapter 3 covers the Pilot study which is an extension of the previous phase in order to assess the feasibility of implementing the measures identified by the HACCP approach through an intervention at household level.

Chapter 4 discusses the results of the study as whole, assessing their validity and their relevance compared to similar results of studies mentioned in the literature review.

The fifth and final chapter draws lessons learnt for the design of hygiene promotion strategies to prevent food borne diseases and so to alleviate the burden of such diseases, mainly among children in developing countries. The thesis concludes with recommendations on research and on how to improve our findings and make them more valuable.

CHAPTER II: HACCP experiment

This chapter describes how the HACCP approach, as described in section 1.3 of Chapter 1, has been applied step by step, to two selected weaning foods prepared by 15 volunteer mothers in peri-urban Mali. This was done on the basis of the findings and recommendations of certain studies (Ehiri et al 2001, Sheth et al 2000) and international food safety strategic papers (WHO, 1999; Bryan, 1992)

After setting the CCPs, actions were taken to control, reduce or eliminate microbial growth at these points using lessons learnt from the five preliminary steps of the HACCP analysis of the modes and sources of contamination of food and relevant measures to prevent them.

For the hazard analysis, and then to assess the efficacy of these measures, 432 food samples were collected from 15 volunteer households, and analysed by the National Laboratory of Public Health in Bamako. Monitoring and verification were applied to the preparation of one meal of each of the two selected foods by each of the 15 mothers.

The experimental phase started in April 2007 and ended in December 2007.

2.1. Assembling the team and definition of the scope of the experiment and action plan

In accordance with the study scope, its objectives and planning, and in accordance with the HACCP practice and with respect to local customs and traditions, I selected three female specialists: a medical doctor specialised in public health, a microbiologist and an agronomist, to contribute to the conception and implementation of the study. Three female field workers with university degrees were also recruited to collect data at household level.

The team had its first meeting in order to agree upon the content of the protocol and to clarify the role of each member. At the second meeting, we discussed selected activities regarding the aim of the study and planned their implementation.

During a third meeting the team presented the proposal to a group of two external HACCP experts chosen to monitor later the implementation of the approach regarding its principles.



Figure 2.1: The HACCP Team

2.2. Description of the product and identification of its intended use

2.2.1. Selection of participants

For the purpose of this study many steps were needed to select volunteer mothers of children aged six to eighteen months. The multistage technique was used to select the study sample as follows. Before contacting households, several meetings were held between the research team, local authorities and community representatives.

The first meeting was held with the mayor and local leaders of Commune V, where Sabalibougou (the zone of the experimental study) is located. The chairmen of three community health centres of the zone and the representative of the Director of the reference health centre were present. During this meeting we explained the objectives and the outcomes of the study and what we expected from them. At the end of the meeting the participants signed a document showing their agreement.

With the document signed by the local authorities and the consent form prepared, we submitted the Protocol to the National Ethics Committee in order to get the authorization to start the field work.

After getting both the National Ethics and the LSHTM Committee's agreement, 13 April 2007 and 20 September 2007 respectively, we continued the process of sample selection.

The second meeting was held with the director of the Commune's reference health centre, coordinator of the three community health centres in our study area. During this meeting we agreed about the involvement of the community health centres,

where we proposed to seek the list of mothers of children under five years old from which we selected our sample.

Accompanied by the respective chairmen, we visited each community health centre. There, we explained to the doctor or the chief nurse how we would like to get a list of mothers visiting their centres. But we found from these contacts that the list of consultations was not useful for selecting mothers because their addresses were not mentioned, so that it would not be possible to find the households where they were living. Alternatively, we planned to attend the nutritional education sessions held one day each week in each centre.

We divided our team into three groups each composed of one female field worker supervised by one female specialist, in order to visit simultaneously the three community health centres twice a week. Each group attended two sessions in each community health centre.

At the beginning of her normal nutritional education session, the midwife explained the objectives and the outcomes of the study to mothers present and introduced our group. If everything was clear, we asked volunteer mothers to raise their hand and give their name, the name, age and sex of their child, their home address and phone number. By the end of the second visit, we had a list of seventy volunteer mothers of children aged between six and eighteen months.

We located easily the households of all the mothers visiting health centres, based on their description of their home address. In our context, a household is defined as individuals who live in the same house and who normally eat at least one meal together in a day.

We wrote the name of each mother on a piece of paper and put them in a box, from which we drew randomly fifteen persons that we numbered from one to fifteen to constitute our sample. The latter was then split into three groups of five mothers each, taking into account their proximity to one of the three community health centres. Each volunteer mother's group of five was followed by one field worker supervised by one professional of the team for data collection.

2.2.2. Households data collection

At this stage two types of data collection tools were devised and tested before use:

- Questionnaire to collect targeted groups (children, mothers and fathers) characteristics and household socioeconomic status,
- Reference persons, for instance elders interview to clarify or check certain information given in the questionnaire.

Results

Table 2.1: Characteristics of household members

Parameters	No. Reported	
	Children's characteristics	
Age (months)		
5- 10		9
11- 15		4
16- 20		2
Sex		
Male		6
Female		9
	Mothers (N = 15)	Fathers (N = 15)
Age (years)		
15-23	7	0
24-34	7	6
35 +	1	9
Educational level		
No schooling	7	6
Primary school	7	6
Secondary school	1	2
Post secondary school	0	1
Occupation		
Housewife ¹	8	0
Trader	4	4
Artisan	0	6
Civil servant	2	3
Driver	0	2
Student	1	0

¹ They did not earn regular income but most of them sell something on the local market.

Table 2.2: Household income and environmental status

Parameters	No. reported (N=15)
Household income group¹	
Low income	12
Medium income	2
High income	1
Refrigerator	
Available	0
Not available	15
Method of excreta disposal	
Latrine	15
Open defecation	0
Children's faeces observed on the premises	1
Distance between latrine and kitchen²	
> 10 m	8
< 10 m	7
Source of domestic water supply	
Well (for utensils and clothes washing) ³	6
Bore hole	14
Home tap	1
Domestic animals in the yard	
Present	8
Absent	7
Food protection during storage	
Totally Covered	12
Partially covered	3
Kitchen utensils⁴	
Clean	12
Not clean	3
Normal cooking fuel	
Firewood	12
Charcoal	3

¹This classification is made on the basis of Malian National Bureau of Statistics and the Census standards. According to these criteria based on goods ownership (house, engines, electronics,), only one household can be classified in the high income class (a couple of civil servants), two in the medium class and the majority in the low income class (earning less than \$ 340 per year); 56.10% of Mali's population pertain to this latter category.

² Minimum distance permitted between latrine and kitchen by the local environmental health office is 10 m. nearly half of households did not meet this standard.

³ These households used water from a borehole or piped supply for drinking and food preparation.

⁴ Cleanness of utensils was assessed by the presence or absence of visible dirt.

Notice: The presence of domestic animals (sheep, goats, chickens and pets) in household yard was considered as an important source of environmental contamination. This was the case for more than half of the households visited.

Sixty percent of the children were under 11 months and 60% of them were female (Table 2.1). Most children's mothers were young (more than 94% were under 35 years old), approximately 50 % of them were illiterate and without regular income (housewives). Contrary to mothers, 60 % of fathers were aged more than 35. On the other hand most fathers in this sample were manual workers. These latter data are consistent with families' income classifications indicated in Table 2.2. However 20% of fathers had reached secondary school.

Table 2.1 shows a clear view of age unbalance between mothers (14/15, more than 93% were under 34 years old) and fathers (9 about 60% are 35 years old). The same Table displays a balance between mothers' and fathers' literacy level, and shows that on one hand, mothers and fathers are equally involved in trading activities but on the other hand certain activities are exclusively exercised by males or females.

2.2.3. Identification and description of the major meals used as weaning food

During focus groups, each mother listed all meals she gave to her child. For each food listed, recipe, raw materials or ingredients, preparation and handling processes were described. Indications were given on how to get ingredients and raw materials. Behaviours related to foods preparation and handling were also listed.



Figure 2.2: Mothers attending a focus group meeting

Table 2.3: Meals regularly given to children as a weaning food

Type of meal	Number reported
“Moni” (gruel made with local cereals)	10
Fish Soup	10
Household meal	4
Meat Soup	1
Cérélac (manufactured baby food)	3
Fruit	3
Milk	3

Table 2.4: Mothers' behaviour during food preparation and handling

Parameters	No. reported (N=15)
Mothers' self-reported hand washing practices¹	
Before touching food	12
After using toilets	14
Procedure for checking temperature of children's food prior to feeding	
Putting a little portion in the mouth	10
Putting a little portion in the palm of the hand	2
Dipping a finger in the food	1
By observation	2
Means of feeding child	
Feeding bottle	1
Plastic cup	10
Spoon	1
Hand	3

¹ When we observed them, mothers' hand washing techniques were very diverse and did not always follow good hygienic practice. Indeed, they did not always wash both hands with running water and soap; sometimes they washed only one hand; they even forgot washing hands on many occasions after using latrines or cleaning a child's bottom. This diversity explains the large difference between these findings based on mothers' self-report, and the result of the hand washing behavioural study conducted by the National Directorate of Health in Mali; only 15% of that study sample members reported that they wash their hands with soap, (Toure, 2006).

Mothers are using many types of meals as weaning food. But, the two most common meals given as a weaning food are "Moni" and Fish Soup (Table 2.3). Table 2.4 summarises mothers' behaviours reported.

2.2.3.1 "Moni" description

This is the most common meal used as a weaning food. Mothers learn its preparation and its use during health education sessions held weekly in community health centres. Therefore, women are strongly advised to use it as a complementary food at the weaning age.

Called enriched flour, this food is composed by up to six local cereals (millet, sorghum, maize, peanut, soybean, wheat). Cereals are bought weekly at the local market. Mixed cereals are soaked and washed in potable water and dried some hours by exposure to sunshine in the household yard. The dried mixture of cereals is ground in a mortar or a mill to make flour. The flour is sieved to separate grains from the powder. This flour is sometimes cooked before adding extra ingredients such as "monkey bread", fruit from the baobab tree used as a preservative and as a substitute for sugar.

Because of mothers' daily activities, many of them spend most of their daily time away from the household; every weekend they prepare the quantity of flour needed for one week's use. The stock is stored in a recycled manufactured powdered milk can.

Every day, the mother takes a small quantity of the flour, adds water and homogenizes it using a spoon or a wooden stirrer. The liquid is cooked during an average of 22 minutes to become the "Moni". Then, sugar, milk or lemon is added. The meal is divided into two parts; one part is given immediately (after ten minutes of cooling) to the baby and the rest is stored at room temperature for seven to twelve hours and served again, reheated or not, to the baby. The latter is fed with a spoon, a cup or from the utensil containing the meal itself.

2.2.3.2 Fish Soup description

This meal is also widely used as a weaning food for the same reasons as Moni. Indeed, it is also promoted by nurses in community health centres during nutritional education sessions. Contrary to cereals, the materials are purchased daily from the local market for same day consumption.

It is composed of fresh fish and vegetables (tomato, onion, and lettuce). Fish and vegetables are washed separately at home. The vegetables are ground before putting them in a pot containing water and set on the fire. The fish, cut into small pieces, is added. The preparation is then cooked for 30 to 50 minutes. Ingredients are added when the preparation is boiling. The soup is split into two parts. One part is used to feed the baby immediately after ten minutes of cooling. The child is fed with a spoon and the mother's hand (to separate fish from bones). The rest is stored until the afternoon, six to seven hours later.

2.2.4 Foods intended use

The foods intended use is related to the volume of foods prepared and to the susceptibility of the targeted consumers. It was clearly established that these foods were used as weaning foods intended for a very susceptible group, the very young.

2.3. Construction of Flow Diagrams and their on-site confirmation

With the same focus groups mentioned above (Figure 2.3), we discussed the supply of raw materials and ingredients, and the different steps of the process of meal preparation, handling, storage and feeding children. All steps were described in detail to ensure a full understanding of the path followed by the meal from obtaining the produce to feeding the child after preparation and storage.

2.3.1 On-site confirmation of Flow Diagrams

For this step two methods of data collection were used: observations and environmental walk.



Figure 2.3: Fresh fish displayed on the local market

The observation stage was conducted with three volunteer mothers. Each mother individually was asked to perform effectively all the process described above. Each mother was followed by one field worker supervised by an expert from our team. We supplied candidates with money to buy raw materials and ingredients. From the local market (where all mothers reported buying produce) to home, the two members of our team observed and noted everything that happened in terms of behaviour and practice during handling, preparation and storage of the meal and child feeding. The team also observed the supply and the handling of water used to clean utensils and to prepare food.

A visit to each household's surroundings and local markets (where raw products and ingredients were bought) were conducted to identify potential sources of microbiological contamination through water and sanitation facilities monitoring: the existence of latrines, their cleanness and their distance from the kitchen and water facility, the location of the kitchen and its cleanness, water supply and storage systems, wastes management system, and presence or absence of domestic animals.

2.3.2. Results

A. Summary of focus groups discussions and observations

Table 2.5: Steps of “Moni” process and sources of contamination (summary from focus groups and observation)

Steps	Sources of contamination
Purchasing raw cereals ¹	Initial contamination: field, dust, handling, containers.
Washing and soaking of cereals	Contaminated water, utensils, handling, dust.
Grinding	Contaminated mortar or mill, utensils, and handling.
Sieving	Dust, contaminated utensils, and handling.
Adding ingredient (monkey bread)	Contaminated ingredient and handling
Flour drying	Contaminated cloths, dust, flies, domestic animals and handling.
Flour cooking ²	Contaminated utensils and handling.
Flour storage	Contaminated utensils, handling.
Flour mixture with water	Contaminated water, mixing materials (spoon, wooden stirrer), utensils containers, and handling.
Cooking	Contaminated utensils and handling.
Cooling	Contaminated utensils, handling, dust and flies.
Adding ingredient (sugar, lemon)	Contaminated ingredients, and handling.
Feeding child	Contaminated feeding utensils (spoons, cups...), handling and flies.
Meal storage	Contaminated utensils, dust, flies, animals contact, insects, storage (temperature, duration).
Reheating ²	Contaminated utensils, handling.
Feeding child	Contaminated feeding tools (spoons, cups...), handling and flies.

¹None of mothers followed ever bought all the kinds of cereals cited during the product description in focus group sessions. The number and the quality of different cereals used depend on the preference and the financial capability of each mother.

²Not performed during observation sessions contrary to mothers' statement during focus group sessions.

Table 2.6: Steps of **Fish Soup** process and sources of contamination (summary of focus groups and observation)

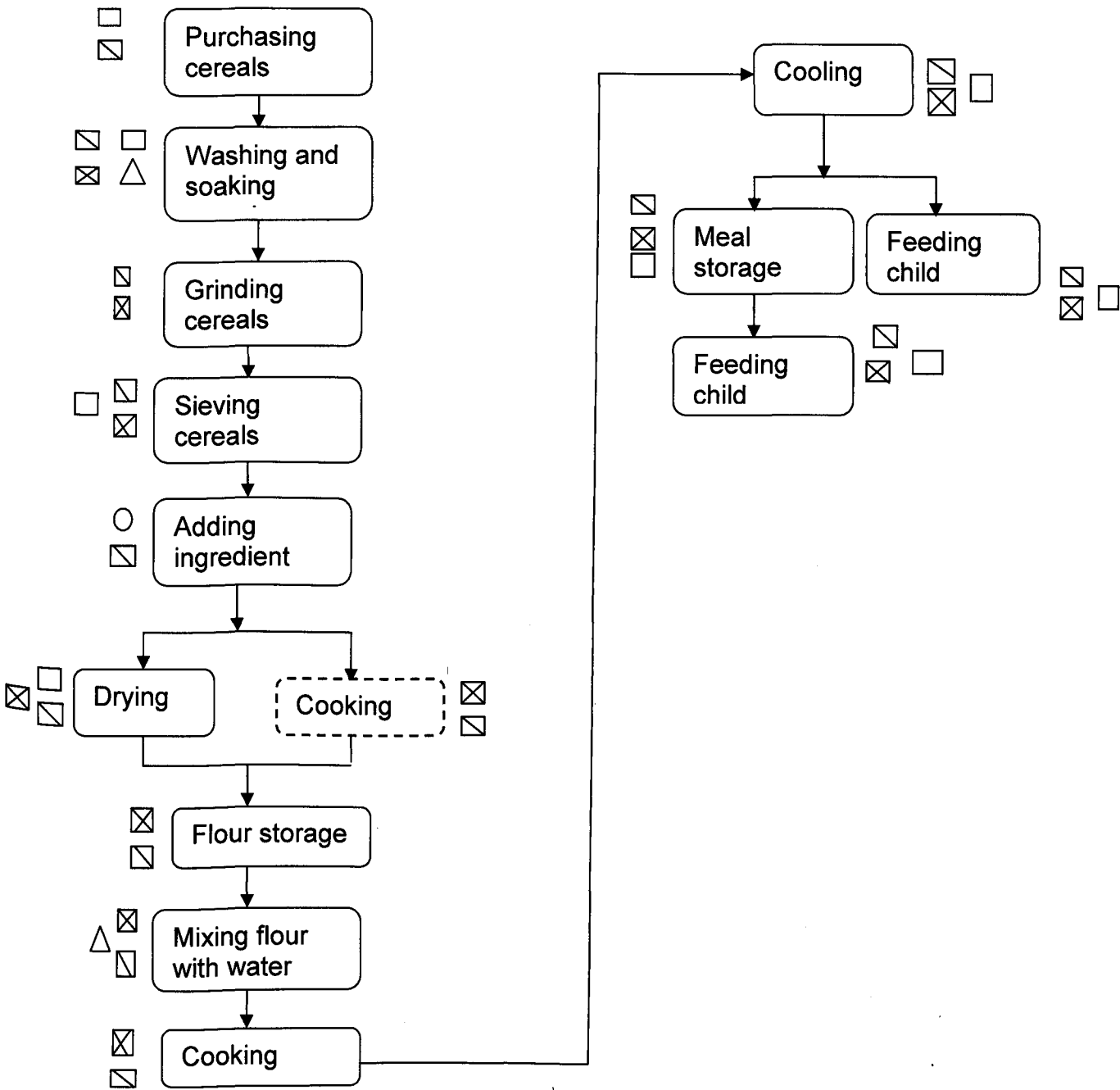
Steps	Source of contamination
Purchasing fish	Initial contamination: from fisheries, dust, handling, containers, knives and flies.
Purchasing vegetables	Initial contamination: contaminated soils, waste water, handling and containers, wrapping materials.
Cleaning/washing fish at home	Contaminated water, knives, utensils, handling and flies.
cleaning/washing vegetables at home	Contaminated water, knives, utensils, handling, flies, and lack of disinfection.
Cooking	Handling and utensils.
Feeding child	Feeding utensils (spoon, cup...), handling, dust and flies.
Meal storage	Contaminated utensils, dust, flies, animals contact, insects, storage (room temperature duration).
Reheating ¹	Contaminated utensils and handling.
Feeding child	Contaminated feeding utensils (spoons, cups...), handling, dust and flies.

¹Not performed during observation sessions, contrary to mothers' statement during focus group sessions.

B. Flow diagrams of meal preparation

Operations/processes are shown in rectangles, each kind of hazard is represented by a symbol and arrows indicate the direction of the flow. Reheating was cited by mothers during interviews, but the step was omitted on the diagrams because it was not actually performed during the observation sessions.

Figure 2.4 : Moni Flow Diagram

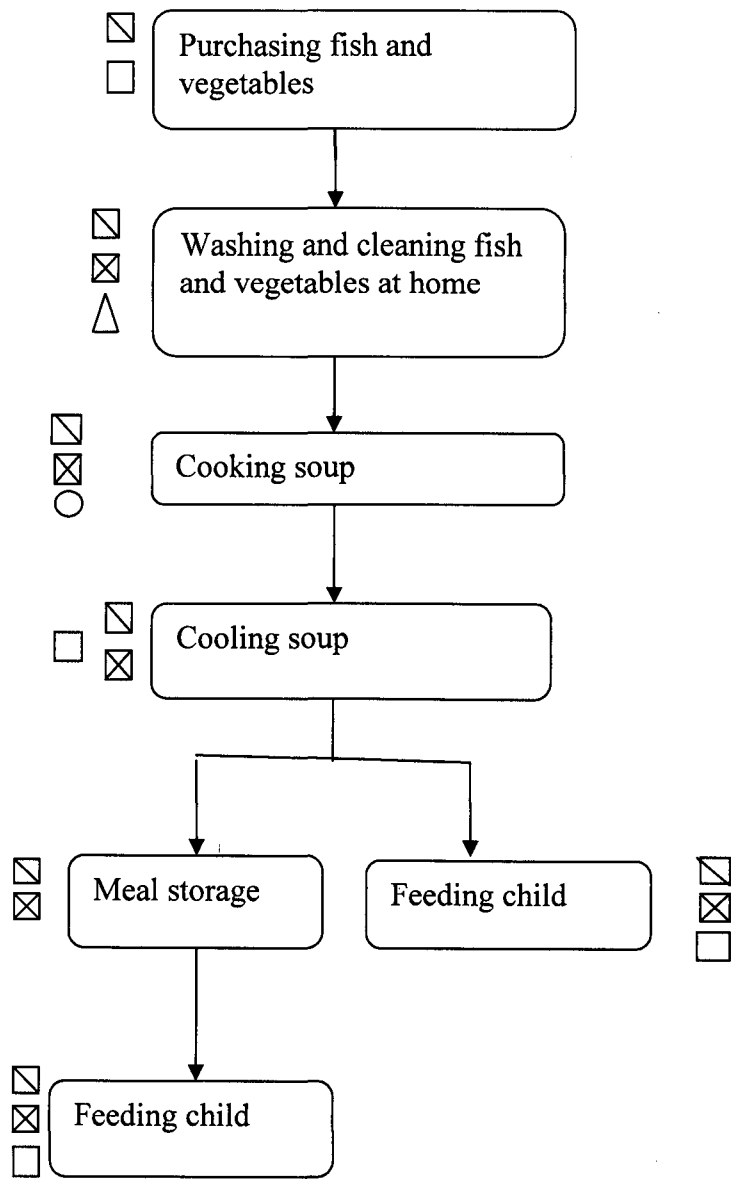


Legend

- Initial contamination
- ▣ Hand contamination
- ▤ Utensils contamination
- Ingredient contamination
- △ Water contamination

--- Cooking not always done

Figure 2.5: Fish Soup flow diagram



Legend

- Initial contamination
- ▣ Hand contamination
- ⊠ Utensils contamination
- Ingredient contamination
- △ Water contamination

“Moni” meal is exposed to diverse sources of contamination from the point of supply (Local Market) to the point of child feeding; and many kinds of contamination can occur at each step of the process (Figure 2.4). Mothers are not always practising what they know. Indeed, flour cooking was not always performed. None of the mothers reheated a meal during our observations, contrary to their statements during focus group sessions. Therefore the step was not included on the flow diagram.

The “Moni” process is very complex; however, the main advantage is that the flour covering the child’s weekly needs can be prepared and stored. Each day a small portion can be quickly and easily prepared for daily feeding. These opportunities render this kind of weaning food very useful for busy mothers.

As observed on Figure 2.4, contamination can also occur at all steps of the Fish Soup meal process (Figure 2.5). For this meal also, reheating did not happen during observation sessions, contrary to mothers’ statements during focus groups meetings. Handling seems to be a permanent source of contamination.

2.4. Hazard Analysis (List of potential hazards and Control Measures)

This step is crucial in the HACCP approach. Indeed, the accuracy with which hazards are identified (and risk of disease associated with them) determine the relevance of actions which must be taken at critical control points to reduce, control or eliminate these hazards.

The step consists of food sampling and measurement of physical parameters at each step of the foods process mentioned on flow diagrams. 432 food samples were taken, examined and used to assess the importance and the severity of FC contamination of foods.

2.4.1. Sampling of Foods

2.4.1.1 Collection of food samples



Figure 2.6: Moni food sampling

Food samples were collected by the National Health Laboratory specialist, assisted by a field worker of our team. Weaning food samples were collected aseptically, before and after cooking, after storage at room temperature (for an average storage time of 8 hours) and immediately after reheating. Raw material samples were obtained from vendors in the local market. Water samples were obtained from community boreholes, vendors, domestic taps, wells and storage vessels. Because of the fact that all mothers declared preparing cereal flour for one week's use, we also sampled flour from each mother's stock.

To collect, to hold and to transport samples, the laboratory technician was equipped with:

- Sterile simple containers: disposal plastic bags (Figure 2.9) for food samples, bottles for water samples, and wrapping papers;
- Sterile and wrapped implements for sample collection: scissors, swabs and sponges;
- Sterile agents: distilled water, propane torch and 95% ethanol;

- Refrigerant: insulated plastic container with ice blocs;
- General equipment: roll of adhesive tape, cotton, soap solution and cloth;
- Clothing: laboratory coat and gloves.

A sample of approximately 200 g was taken using the traditional cup used by the mother to homogenise, cool and feed her child (Figure 2.9). The samples were put in sterile plastic containers with tight-fitting lids. Hot food samples from households were cooled immediately in an insulated plastic transport box containing ice blocks and were kept there, at 4.4 °C, until arrival (after approximately 3 hours) in the Laboratory. The sample was labelled with a code that identified the household (serial number, group and location). It is important to notice that the laboratory technicians were blinded to the nature of food samples.

Before collecting samples the temperature, pH and humidity of the foods were measured. For these parameters measurement the following equipment was used:

- Temperature measurement: Thermometer TFX410-1, Klipspringer Instrument, Rynor House, Farthing Road, Ipswich IP1 5AP, UK, measuring range -50°C to 300°C, resolution +/- 0.1°C, accuracy +/- 0.2°C +/- 1 digit and dimensions 109x54x22 mm;
- pH measurement: Hand Held pH/mv meter model 8601, Klipspringer Instrument, Rynor House, Farthing Road, Ipswich IP1 5AP, UK, measuring range 0.00 to 14.00, resolution 0.01 pH, accuracy +/- 0.02 pH;
- Humidity measurement: Hygrometer TFH 610, Klipspringer Instrument, Rynor House, Farthing Road, Ipswich IP1 5AP, UK, measurement range 0 to 100 % rH, resolution 0.1 % rH, accuracy +/- 2.5 % rH;
- Duration measurement: Chronometer ALBA W071, made in Japan.

Sterile digital thermometers were used to measure meal temperatures at three points: immediately after cooking, after storage at room temperature for an average of 8 hours, and immediately after reheating. Using a pH meter and humidity meter, we measured the pH and the humidity of the flour in mothers' stocks. When the flour was cooked before storage during one week, we measured the temperature immediately after cooking. In total 432 samples were collected and examined. Each food or water sample collected was recorded in the field notebook and labelled at the point of collection.

Given that the significance of a health hazard depends on the likelihood of its occurrence and the severity of its impact on the health of the target group, we considered only bacterial hazards which are of a greater importance in Mali. Indeed the most common and the most severe diarrhoeal disease of young children is largely caused by bacterial hazards of faecal origin (MS/CPS, 2007). Accordingly, one group of faecal indicator bacteria (*faecal coliforms*) had been considered as indicator of Potential Hazards and was counted as measurement of microbiological food quality.

2.4.1.2. Examination of samples

Depending on the capacity of the Laboratory, the samples were either analysed immediately, or kept in a refrigerator (at 4 degrees Celsius), till their examination no later than 72 hours.

To test for the presence of FC, the Enumeration of thermotolerant coliforms by colony-count technique at 44°C – Routine method was used (French Association of Standardization, AFNOR, NFV 08-060, March 1996) .

Dilution: buffered peptone water was used.

Medium: Violet Red Bile Lactose Agar (VRBL).

a) Food

• Dilution procedures

10 g of food was taken from the 200 g sampled in households and diluted in 90 ml of buffered peptone water and homogenised using a mixer bag. The solution was decanted during 15 minutes (solution 10^{-1}). 1 ml of the supernatant was transferred in a tube containing 9 ml of sterile solution and mixed (solution 10^{-2}).

• Culture techniques

1 ml of the solution (solution 10^{-2}) was poured onto a sterile plate, and 15 ml of medium was added. The plate content was perfectly homogenised and let to solidify before incubation at 44 °C for 24 hours.

• Colonies count

Violet colonies circled with red zone with a diameter larger than 0.5 mm were counted. Only plate containing less than 150 characteristic colonies were considered and each plate must contain at least 15 characteristic colonies. The number N of coliform thermo tolerant per gram of food is given by the following formula:

$$N(\text{coliform thermotolerant per gram}) = \frac{\sum C}{1.1 \times d}$$

- $\sum C$, is total of characteristic colonies counted on the two successive plates;
- d, is the dilution factor corresponding to the first dilution ($d = 10^{-1}$ in our case).

If the plate of the test sample contained less than 15 characteristic colonies, the following formula were used

$$N = a/d$$

- a is the number of characteristic colonies counted
- d is the dilution factor

If the plate of the test sample contained no characteristic colony

$$N = 1 \times 1/d$$

b) Water

• Culture techniques

1 ml of water was poured onto a sterile plate and 15 ml of the medium was added. The plate content was perfectly homogenised and let to solidify before incubation at 44 °C for 24 hours.

• Colonies count

It is similar to that presented in food, however, if characteristic colonies counted are less than 15, N (Coliform thermotolerant per ml)= a, where a is the number of characteristic colonies counted on the test plate; N < 1 Coliform if no characteristic colony was counted on the test plate.

2.4.2. Results

Table 2.7: Physical parameters (means) of cereals and monkey bread

Materials N= 5 for each	Parameters	
	Temperature(°C)	Humidity (%)
Maize	31.4	68
Rice	34.5	69
Sorghum	33.7	60
Millet	44.8	63
Monkey bread	32.8	50

Table 2.8: Physical parameters (means) of the flour and weaning foods

Food	Parameters			
	pH	Humidity	Temperature °C	Duration cooking/storage
Flour		51,4	29.5	1week storage
"Moni" after cooking	4.3	-	91.1	22 min cooking
"Moni" after storage	4.5	-	36.6	More than 6 hours storage
Fish soup after cooking	4.9		87.8	40 min cooking

Fish soup after storage	5.8	-	33.2	More than 6 hours storage
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Table 2.9: Water, raw products and ingredients FC count

Item	<i>Faecal coliform</i> count per 100 ml (liquids) or per gram (solids)	N° of samples (15 households)
Water		
Well	20-60	5
House connection	< 1	2
Borehole	< 1	2
Jar	< 1	2
Vendor	50-160	7
Container	60-310	11
Solids		
Maize	< 1	2
Rice	20-60	2
Sorghum	20-60	2
Millet	100-120	2
M. bread	< 1	2
Flour	50-1300	15
Fresh. fish	110-840	15
Vegetables	20-1000	15

Table 2.10: FC count (FC/g) in Moni before and after cooking

H.H Reference	FC count per gram		
	Before cooking	Immediately after cooking	After storage
1	170	ND	ND
2	330	ND	ND
3	210	ND	290
4	210	ND	ND
5	370	ND	ND
6	1000	ND	120
7	1600	ND	ND
8	500	ND	ND
9	70	ND	ND
10	150	ND	ND
11	280	ND	ND
12	130	ND	ND
13	130	ND	ND
14	590	ND	ND
15	180	ND	ND

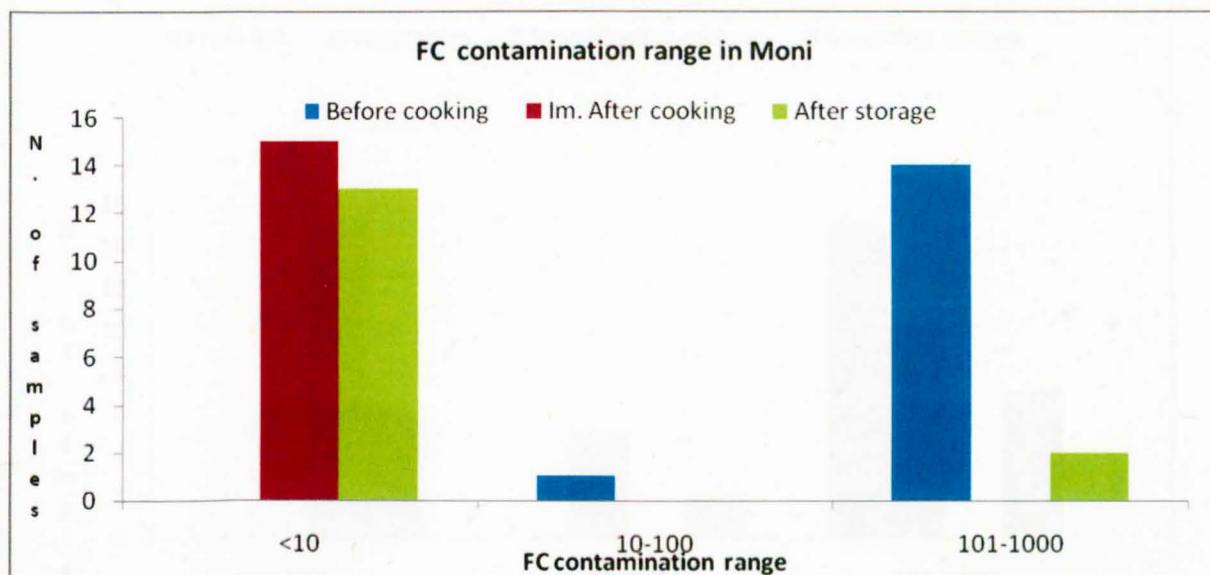


Figure 2.7: FC contamination range (FC/g) in Moni before and after cooking

Table 2.11: FC count (FC/g) in Fish Soup and its raw products (Fresh fish and vegetables)

H.H Reference	FC count per gram			
	Before cooking		Immediately after cooking	After storage
	Fresh fish ¹	Vegetables ¹		
1	190	80	ND	190
2	190	40	ND	160
3	120	78	ND	ND
4	350	20	ND	510
5	840	150	ND	ND
6	180	400	ND	30
7	130	820	ND	110
8	190	1000	ND	100
9	190	1000	ND	550
10	410	230	ND	ND
11	550	160	ND	ND
12	170	290	ND	ND
13	190	150	ND	ND
14	570	240	ND	120
15	110	20	ND	150

¹ Fresh fish and vegetables are actually raw products which are later mixed and cooked to make soup.

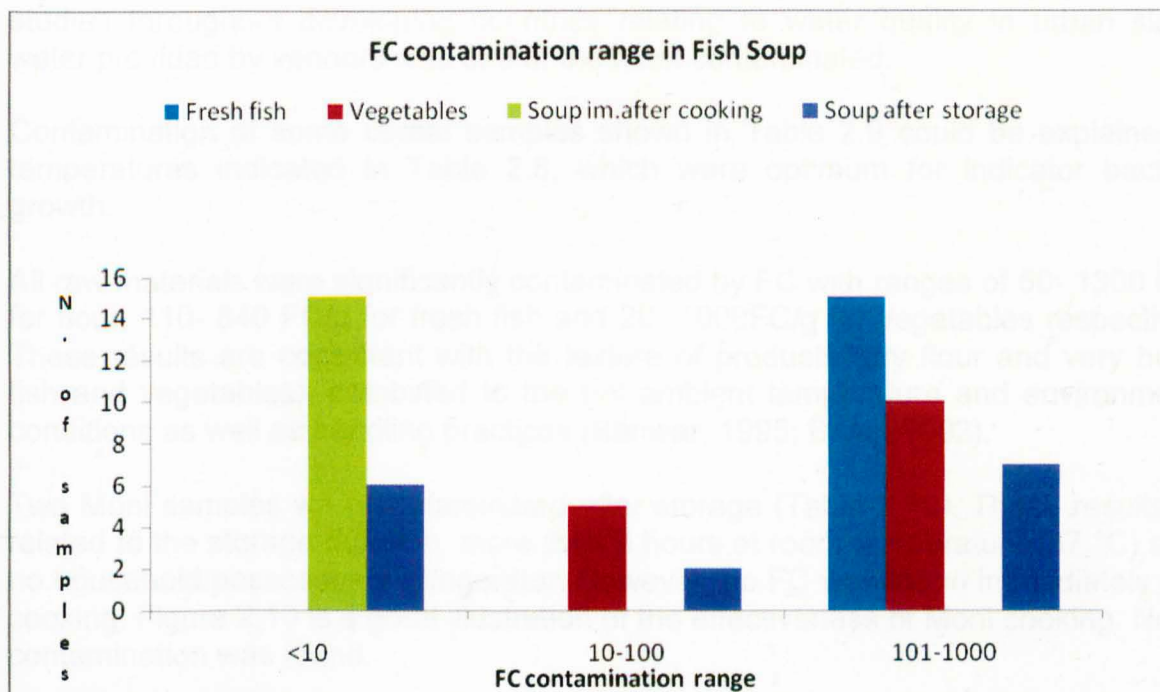


Figure 2.8: FC contamination range (FC/g) in Fish Soup and its raw products (Fresh fish and vegetables)

Raw product temperatures correspond to the most suitable temperatures for pathogen growth (31.4-44.8 °C) (Table 2.7), but humidity is less than that required to allow the multiplication of those species (Bryan, 1992; Banwar, 1995).

Values of pH in the range 4.8- 5.8 are the minimum for the growth of most of food-borne pathogenic bacteria (Bryan, 1992); the temperatures of stored foods and storage duration are the most suitable for pathogens growth, making storage the most harmful step in the whole process of foods preparation and handling (Table 2.8). On the other hand, cooking temperatures are far higher than temperatures necessary (60°C) for FC destruction (Banwart, 1995; Griffith, 1994, 2003). That is confirmed by bacterial analysis of all cooked food samples (Tables 2.10 and 2.11). Indeed no FC were found in either Moni or Fish soup at this stage of their process.

Water

House connections and Boreholes were free of FC; these results are consistent with weekly water quality monitoring data of the Bamako urban water supply system, published by the Water Quality Laboratory.

For the other water sources, samples were taken from any kind of reservoir found in the household. Many sources were found in the same household e.g. jar for water storage inside living rooms, or container shared by several households living in a compound in which a traditional well is also present. Vendors were selected from a group getting water from private boreholes and samples were collected when they were delivering water in households.

Unsurprisingly, water from traditional wells was the most contaminated, followed by containers filled with water from a piped house or borehole. As found in several

studies throughout developing countries relating to water quality in urban slums, water provided by vendors was one of the most contaminated.

Contamination of some cereal samples shown in Table 2.9 could be explained by temperatures indicated in Table 2.8, which were optimum for indicator bacterial growth.

All raw materials were significantly contaminated by FC with ranges of 50- 1300 FC/g for flour, 110- 840 FC/g for fresh fish and 20- 1000FC/g for vegetables respectively. These results are consistent with the texture of products (dry flour and very humid fish and vegetables), combined to the hot ambient temperature and environmental conditions as well as handling practices (Banwar, 1995; Bryan,1992).

Two Moni samples were contaminated after storage (Table 2.10). These results are related to the storage duration, more than 6 hours at room temperature (37 °C) since no household possesses a refrigerator. However, no FC was found immediately after cooking. Figure 2.10 is a good illustration of the effectiveness of Moni cooking. No FC contamination was found.

Most Fish Soup samples were contaminated after storage (Table 2.11). These results are related to storage for more than 6 hours at ambient temperature since no household owns a refrigerator. Additionally the two raw components of this meal were highly contaminated making it more vulnerable. However, no FC was found immediately after cooking. Figure 2.11 confirms that cooking is as effective at FC elimination in Fish Soup as in Moni.

Tables 2.10 and 2.11 show that traditional cooking is very effective at FC destruction in both Moni and Fish Soup. This result is due to the high temperature (87- 91°C) of cooking and its long duration (22- 40 min) (Table 2.8) which are far higher than critical criteria (60°C during 12 min) needed for pathogen elimination (Banwart 1995; Pawsey 2002; Griffith 1994,2003).

2.5. Determine Critical Control Points (CCP)

A CCP is an operation (practice, procedure, location or process) at which control can be exercised over one or more factors to eliminate prevent or reduce a hazard.

2.5.1. Selection of CCPs

For the determination of the CCP, each process step of the food flow diagram was verified as to whether it satisfied the following questions from the decision tree (Figure 2.1) (answering by Yes or No).

Question 1: *Do preventive control measures exist?*

Question 2: *Is the step specifically designed to eliminate or reduce the likely occurrence of a hazard to an acceptable level?*

When a process step satisfied both Questions 1 and 2 (two questions answer was yes), the step was considered as CCP,

If a step only satisfied one question (question 1 answer was yes but question two answer was no), subsequent questions would be asked.

Question 3: *Could contamination with identified hazard(s) occur in excess of acceptable level(s) or could this increase to unacceptable levels?*

Question 4: *Will a subsequent step eliminate identified hazard(s) or reduce likely occurrence to acceptable levels?*

When a process step satisfied question 3 (question 3 answer was yes) and did not satisfy question 4 (question 4 answer was no), the step was also considered as CCP.

When even question 1 was not satisfied (question 1 answer was no), the subsequent question would be:

Question: *Is control at this step necessary for safety?*

When the answer of this question was yes, then the step was modified to introduce a CCP.

Applying the decision tree instructions as described above, it appears that *cooking* is the first CCP for both Moni and Fish Soup. Indeed, cooking is an effective control measure of microbial growth and the step is specifically designed to eliminate bacterial hazards (Tables 2.10 and 2.11). Cooling cooked food before child feeding is also a CCP because bacterial contamination could occur in excess, depending on handling conditions and duration of cooling, and no subsequent step exists to control a hazard.

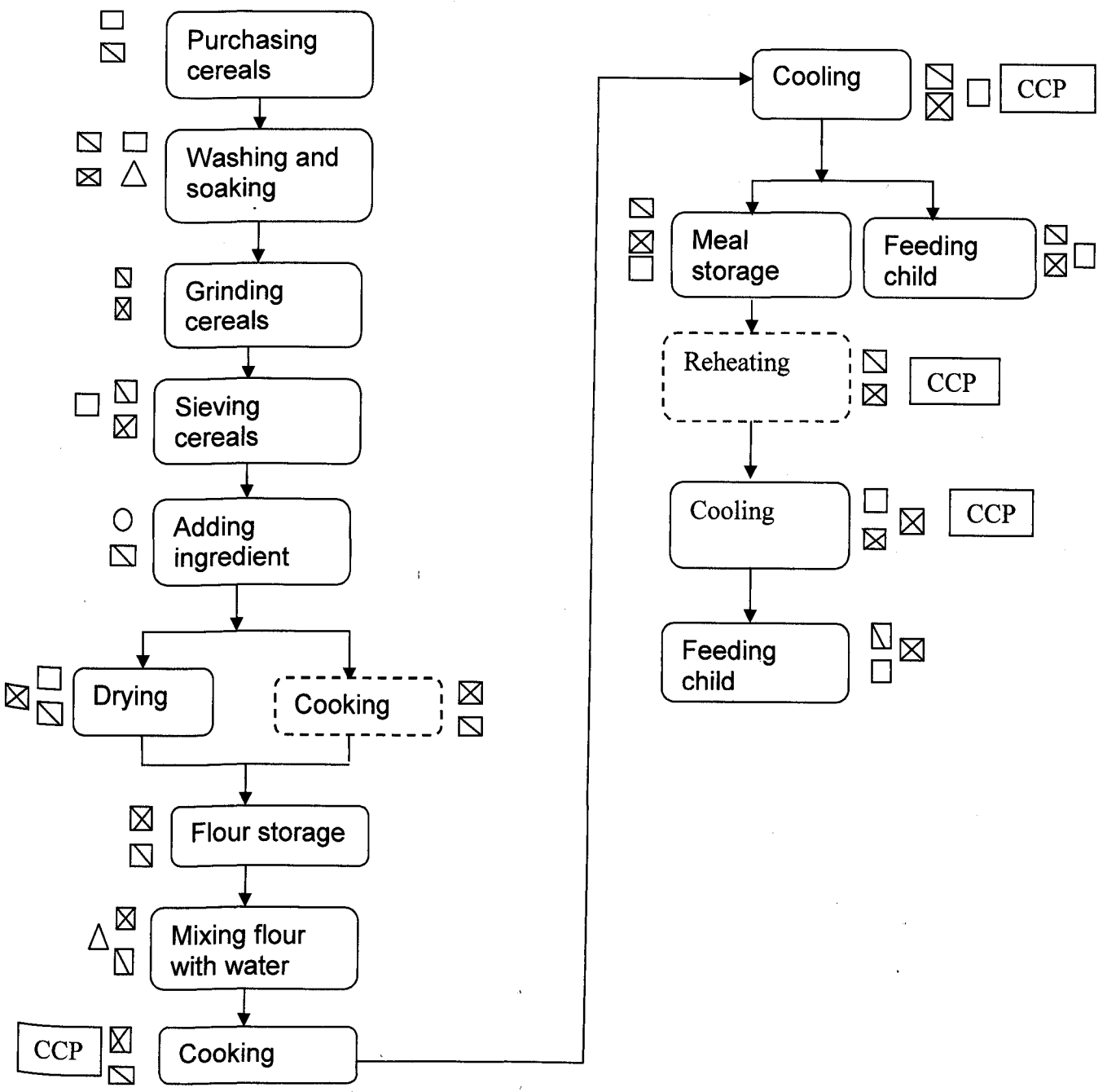
Food stored during more than six hours at room temperature and then served to children is the most hazardous possibility of all, as highlighted in Tables 2.10 and 2.11. Not only are FC found at an unacceptable level but also, no subsequent step exists to eliminate or reduce FC to a acceptable level. Thus it is necessary to add a new operation to reduce faecal contamination prior to child feeding. Our team decided therefore to introduce leftover reheating in the process of preparation of two of the foods. The challenge became to find suitable messages to convince very busy and poor mothers about the relevance and feasibility of this measure in protecting their children's health so that they will adopt it.

All operations occurring before cooking, in both Moni and Fish Soup, are not considered as CCP because cooking as a subsequent step is effective to eliminate FC.

The application of the HACCP principles produces the following diagram showing the selected CCPs.

2.5.2. CCP Diagrams for the two Foods

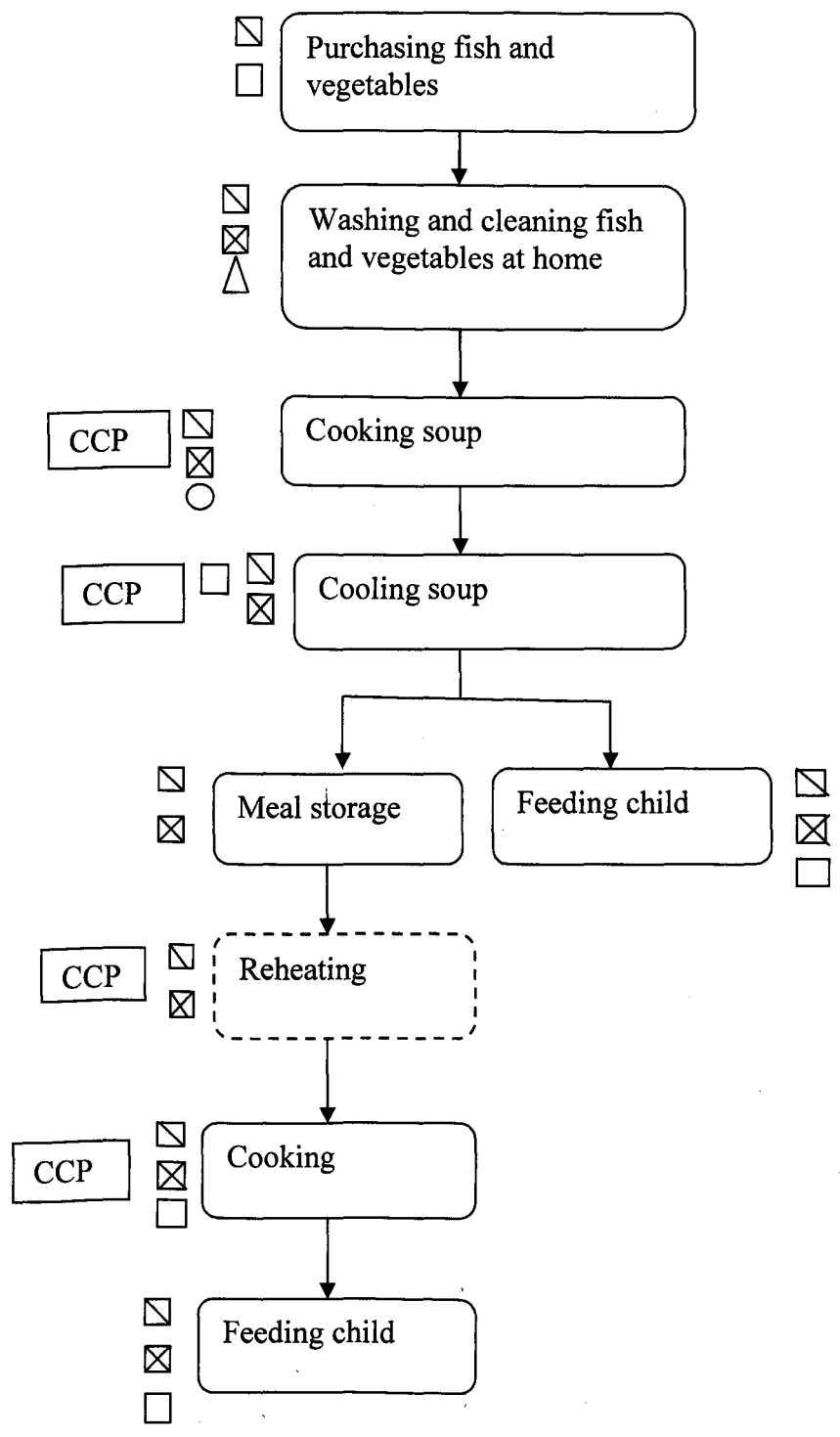
Figure 2.9 : CCP of Moni process



Legend

- Initial contamination
- Hand contamination
- Utensils contamination
- Ingredient contamination
- Water contamination
- Reheating new process introduced by HACCP
- Cooking not always happened

Figure 2.10: CCP of Fish soup process



- Legend**
- Initial contamination
 - ▣ Hand contamination
 - ⊠ Utensils contamination
 - Ingredient contamination
 - △ Water contamination
 - - - Reheating new process introduced by HACCP

For both “Moni” and Fish Soup, the decision tree scheme led to the identification of two CCPs: cooking and reheating. However, considering the child’s exposure to hazard, the moment of their feeding with cooled food, whether after cooking or reheating, is a point at which pathogens are potentially ingested, putting child at risk. Indeed, after cooking or reheating, the process of cooling exposes meals to contamination from the mother’s handling, utensils, flies, domestic animals contact, and dust. Therefore, control of microbial growth at these points is necessary for safety; then, control measures must be implemented justifying the introduction of CCPs at these steps.

In total, for each weaning food, we found 4 CCPs: (i) **cooking**, (ii) **reheating after storage**, (iii) **cooling before child feeding with cooked meal** and (iv) **cooling before child feeding with reheated meal**.

2.6. Establishing critical limits for each CCP

After identification of CCPs, applicable control measures must be implemented to ensure the safety of the selected foods. These measures must be practicable and economically feasible.

2.6.1. Physical criteria

It is obvious that, at all CCPs selected, temperature and time are the relevant factors of microbial destruction or growth. For cooking for example, the higher the temperature, the more effective is the destruction of pathogens for a given time; and the longer the duration of high temperature, the more complete is the elimination of germs. Accordingly, the couple temperature- time has been chosen for cooking and reheating. Critical limits selected are (70°C – 10 min), as (60°C - 12 min) is enough to eliminate pathogens (NSF, 2006; Reinhold, 1995).

2.6.2. Behavioural criteria

Preventing foods from recontamination after cooking and reheating during cooling time prior to child feeding, requires mothers or care-givers to change their behaviour and keep everything in touch with foods free of microbiological hazards. It implies using clean utensils and clean hands to handle foods. Accordingly, criteria selected were:

- hand washing with safe running water and soap after using latrines or cleaning a child’s bottom; and before food handling and child feeding,
- Washing utensils with soap and rinsing them with safe running water.

2.7. Monitoring system for CCPs

The aim of monitoring is to detect any deviation from established criteria. Monitoring of CCPs is essential to ensure that the specified criteria are being met. Results must be obtained immediately so that corrective actions can be quickly taken.

2.7.1. Monitoring procedures for the selected weaning foods

On the eve of each visit, an amount of money (1000 CFA), approximately one pound, was given to the candidate mother in order to buy raw materials of the meal scheduled. If Moni was the proposed meal, raw materials were transformed into flour just after their purchasing.

At nine o'clock in the morning, the field worker arrived in the household equipped with a thermometer, a chronometer, a pH-meter and a form for data collection. The field worker followed the mother at each step of the process of the scheduled weaning food preparation to make sure that all corrective actions were implemented as indicated.

Utensils were always carried to the tap or to the borehole area for washing. The mother and the field worker washed utensils with running water and soap. Water for drinking and food preparation was fetched and carried home.

The candidate mother informed the field worker when she started cooking and reheating. The field worker switched on the chronometer and waited until the mother considered the meal was well cooked; then the chronometer was switched off and the time recorded. The meal became ready to be taken off the fire and cooled. At any time when the mother performed a risky behaviour (for example, touching a contaminated object or cleaning a child or using the latrine), she immediately washed her hands with running water and soap before resuming the handling of food. The session ended around five o'clock in the afternoon.

2.7.2. Monitoring results for selected weaning foods

Table 2.12: Means, maxima and minima of cooking and reheating temperatures recorded (°C). N = 15

	Moni		Fish Soup	
	Cooking	Reheating	Cooking	Reheating
Mean	98.3	97.4	98.98	96.24
Max	99.0	98.7	99.3	99.7
Min	94.1	86.7	98.5	77.8

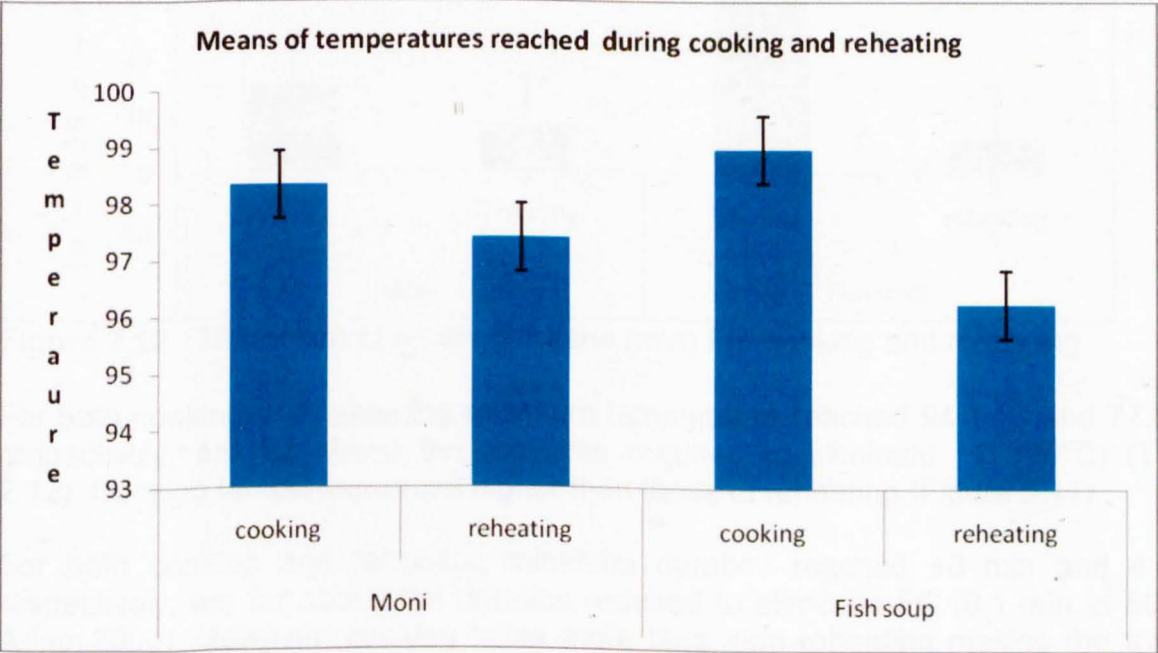


Figure 2.11: Means of temperatures reached during cooking and reheating for each food

Table 2.13: Means, maxima and minima of duration (min) for cooking and reheating=15

	Moni		Fish Soup	
	Cooking	Reheating	Cooking	Reheating
Mean	15.1	7.7	44.3	5.9
Max	20	11	65	9
Min	10	4	25	4

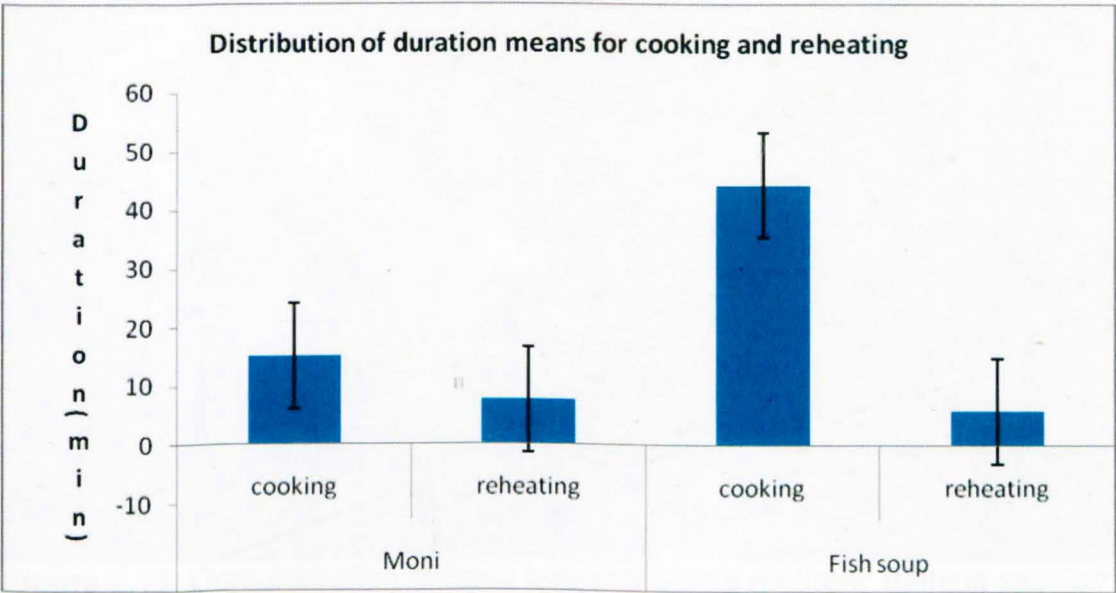


Figure 2.12: Distribution of duration means (min) For cooking and reheating

For both cooking and reheating minimum temperature reached 94.1 °C and 77.8 °C respectively, are far above temperatures required to eliminate FC (60°C) (Table 2.12). Cooking temperatures are higher than those of reheating (Figure 2.11).

For both cooking and reheating minimum duration reached 10 min and 4 min respectively, are far above the duration required to eliminate FC (0.1 min at 60° C; Adam,2000). However, cooking takes more time than reheating making the former operation safer (Table 2.13). Reheating time is shorter than cooking time.

2.8. Establishing Corrective Actions



Figure 2.13: Demonstration of hand washing during mothers' training session

Corrective actions must be immediately taken when monitoring indicates that a process is out of control or that criteria are not being met. One of the positive features of the HACCP approach is that unacceptable contamination, or the existence of a condition that would permit multiplication of undesirable microorganisms can be detected as it occurs, so that immediate corrective action can be taken.

As indicated in Section 2.2.1 (participants' selection), the 15 households of the sample were divided into three groups of five (5) households on the basis of their proximity to one of the three community health centres. Each group was under the responsibility of one female field worker supervised by one female professional of the team.

As reheating was not mothers' habitual behaviour, it became necessary to think about how to convince them to adopt it in addition to cooking. Thus, the team listed all unhygienic behaviours registered during observational sessions (Tables 2.5 and 2.6).

Discussions were opened and participants exchanged ideas on simple measures to improve food safety through behavioural change. The following corrective measures were selected:

- Washing hands with safe running water and soap (including local soap), at the critical periods (before starting meal preparation, feeding children or eating, after cleaning a child's bottom and after using latrines),
- Washing dishes with safe running water and soap (including local soap), using safe water for the preparation of food as well,
- Cooking and reheating foods until boiling for a moment, and
- Covering foods hermetically during storage.

In accordance with the hazard analysis results (Table 2.9), only water from a domestic tap or borehole was regarded as safe and to be used for all domestic needs. Accordingly mothers were advised to fetch water from a tap or borehole and use it immediately, to avoid a long period of storage.

The three female field workers were trained in the implementing and the monitoring of these corrective measures. Instead of giving to each household a garbage container, as planned in the study protocol, as a compensation for their involvement, we decided to provide them with hand washing equipment (kettle and basin) (Figure 2.13) and local soap.

Each field worker held a meeting with the five mothers of the group and presented hygienic outcomes (Tables 2.7-2.11) and proposed measures to tackle unhygienic behaviours selected.

Mothers' training included practising the behaviour changes needed, mainly a demonstration of effective hand washing practice (Figure 2.13). Each field worker agreed with her group members upon a schedule of household visits. Verification activities started three weeks later.

2.9. Verification Procedures

Verification is the step to assess the appropriateness of control criteria and CCPs. It must be undertaken by an independent and qualified person.

2.9.1. Verification method

As FC contamination is the only hazard considered in this experiment, food sampling for the FC count is retained to assess the effectiveness of the approach. Indeed the sole objective is to find out how far FC micro organisms have been reduced or eliminated in the two selected weaning foods using the HACCP approach. 30 food samples were taken at CCPs (cooking, reheating and cooling prior to child feeding), 15 samples for each food. Sample collection and examination were carried out by the National Public Laboratory specialist in the same conditions as in the hazard analysis step.

2.9.2. Results

Table 2.14: FC count (FC/g) for quality evaluation in Moni

H.H Reference	FC / g		
	After cooking	Cooled food after cooking	After reheating
1	ND	ND	ND
2	ND	ND	ND
3	ND	ND	ND
4	ND	ND	ND
5	ND	ND	ND
6	ND	ND	ND
7	ND	ND	ND
8	ND	ND	ND
9	ND	ND	ND
10	ND	ND	ND
11	ND	ND	ND
12	ND	ND	ND
13	ND	ND	ND
14	ND	ND	ND
15	ND	ND	ND
% Positive	0%	0%	0%

Table 2.15: FC count (FC/g) for quality evaluation in Fish Soup

H.H Reference	FC / g		
	After cooking	Cooled food after cooking	After reheating
1	ND	ND	ND
2	ND	ND	ND
3	ND	ND	ND
4	ND	ND	ND
5	ND	ND	ND
6	ND	ND	ND
7	ND	ND	ND
8	ND	ND	ND
9	ND	ND	ND
10	ND	1000	ND
11	ND	ND	ND
12	ND	ND	ND
13	ND	72	ND
14	ND	ND	ND
15	ND	ND	ND
% Positive	0%	13%	0%

Table 2.14 above shows that the Moni was free of FC at all steps of the process and in all samples. The growth of bacterial observed in two samples (290 and 120 FC/g respectively) during storage in bacteriological analyses before the experiment (Table 2.11), did not happen; presumably, the bacteriological quality improvement at this step could be ascribed to the hygienic corrective measures implemented (hand washing etc.).

It appears also that hygiene measures were very effective in controlling FC in fish soup during storage. Indeed, 9 out of 15 samples were FC contaminated (range: 120- 550 FC/ gram) before the experiment (Table 2.11), but only two samples are found contaminated after the experiment (72-1000 FC/g) (Table 2.15). In these latter cases, field workers' observational records showed that preventive measures had not been properly implemented by the mothers concerned. Indeed, one of them never washed her hands prior to food handling and the other washed her hands occasionally after using the latrine.

2.10. Establishing Documentation and record keeping

All data collected during the study were recorded on computer files and papers.

2.11. Data analysis

Table 2.16: Comparison of physical and bacteriological parameters of two weaning foods; P values relate to the null hypothesis of no difference between raw material and prepared foods

Foods and raw materials	Means	P NS– not significant
FC count (\log_{10} FC/g)		
Moni before cooking	2.44	0.004
Moni after storage	0.30	
Moni after cooking	0.00	0.201
Moni after storage	0.30	NS
Moni before cooking	2.44	0.002
Moni after cooking	0.00	
Fresh fish	2.38	0.850
Vegetables	2.22	NS
Fresh fish	2.38	0.030
Fish soup after storage	1.31	
Vegetables	2.22	0.080
Fish soup after storage	1.31	NS
Temperature		
Moni cooking	98.30	0.261
Moni reheating	97.40	NS
Fish soup cooking	98.98	0.071
Fish soup reheating	96.24	NS
Cooking/Reheating duration		
Moni cooking	15.10	< 0.0001
Moni reheating	7.70	
Fish soup cooking	44.30	< 0.0001
Fish soup reheating	5.90	

There is significant difference in FC contamination levels between cooked and stored Moni, between raw and cooked Moni and between fresh fish and Fish Soup after storage. No difference appears between cooked and stored Moni, between fresh fish and vegetables, and between cooked and reheated fish Soup.

No significant difference is perceptible between cooking and reheating temperatures in Moni and Fish Soup, but there is a very significant difference between cooking and reheating durations in both Moni and Fish Soup.

2.12. Discussion and lessons learnt

Hazard analysis demonstrated that the selected foods were faecally contaminated at all stages of their handling (Tables 2.9 to 2.11). This contamination was due to many sources described in Tables 2.5 and 2.6.

For each of the two weaning foods considered, four Critical Control Points (CCPs) were identified: cooking, reheating and cooling after cooking and after reheating. Ehiri (2001), in the study of Critical Control Points of complementary food preparation and handling in eastern Nigeria, identified 5 CCP, including reheating, for the akumu preparation and handling, and Sheth et al (2000), in the study of Hazard Analysis and Critical Control Points of weaning foods, selected two CCP: roasting and storage.

During the whole process of preparation and handling of foods, no sample taken immediately after cooking was found to be contaminated (Tables 10-11). Therefore, mothers' traditional cooking appears to be very effective for pathogen elimination.

Storage was reported as the most hazardous step in the whole process due to storage at ambient temperature in tropical regions, about 33- 37 °C (Table 2.8), the most suitable temperature for pathogen growth (Bryan, 1992, Banwart, 1995) and its duration, at least 6 hours (Table 2.8), far longer than the 90 minutes recommended in these conditions (Bryan, 1992, Griffith, 1994).

Reheating, the corrective action implemented to control microbial growth after storage, is not a traditional behaviour of mothers in our study sample, but appeared to be very relevant for pathogen elimination, because temperatures reached, at least 77°C (Table 2.12) and duration, at least 22 min (table 2.13) were greater than required for these organisms' destruction, namely 60 °C and 12 min, (Bryan, 1992, Banwart, 1995). In addition there is no difference between cooking and reheating temperatures ($P < 0.0001$) (Table 2.14). It could be concluded that reheating is as effective as cooking.

Both Moni and Fish Soup are subject to the risk of microbiological faecal contamination during cooling prior to child feeding, even if all microbes were supposed to be eliminated at the cooking stage. This could be linked to cross-contamination from handling in a very hazardous environment, (Tables 2.5 and 2.6) and confirmed by hazard analysis, (Tables 2.9 to 2.11). Corrective measures implemented at these steps: (hand washing at critical periods and cleaning utensils with potable water and soap), seem to be effective mainly for the Moni meal.

In conclusion, the HACCP experiment significantly improved the bacterial safety of the two weaning foods considered.

Corrective measures/actions which appeared to be effective for controlling hazards at Critical Points for the two weaning foods were:

- Cooking and reheating foods at temperature and duration greater than those of the critical limits,
- using clean water for household needs (drinking, cooking washing dishes...): water from home tap or community borehole,
- washing hands with clean water and soap at critical moments: : after using latrines or cleaning a child's bottom, and before eating, food handling and child feeding,

The actions listed above were translated into educational messages for the following (pilot) phase aiming to confirm the effectiveness of the HACCP approach in improving food safety at household level.

CHAPTER III: Pilot study

3.1. Introduction

The HACCP experimental study (Chapter 2) has shown that, the implementation of the HACCP approach could improve the microbiological safety of weaning food. But the question is: Could messages delivered to mothers about corrective actions to be taken at Critical Control Points (CCP) result in microbial reduction in weaning foods?

The pilot study described in this Chapter aimed to find out an answer to this question. In the experiment, four CCPs: (i) cooking, (ii) reheating, (iii) cooling after cooking, and (iv) cooling after reheating, were identified for each food. Four actions appeared also to be relevant to control bacterial hazards at these CCPs: (i) cooking food thoroughly, (ii) reheating food thoroughly, (iii) hand washing with clean water and soap (including local soap) at critical periods (after using latrines and cleaning a child's bottom, and before touching foods) and (iv) using clean water and soap (including local soap) to wash dishes. Water is regarded as clean when fetched from a stand-pipe or borehole and used immediately.

The sample of 60 volunteer mothers selected randomly were split into two groups of 30; the first underwent the intervention (delivery of messages directed to the actions above) and the second set as a control.

For the pilot study we retained the same two foods (Moni and Fish Soup) which were studied during the experiment and whose CCPs were well established allowing us to start the implementation of actions to improve foods microbiological safety through monitoring of CCPs.

Food samples were taken for bacterial examination. Simultaneously physical parameters were measured, as in the experiment, in 60 households before the intervention. These initial data were used as baseline. Then the Intervention Group underwent training and demonstrations to enable each member to perform actions to improve her child's food.

The three female field workers recruited during the experiment continued to follow mothers. Each of them was in charge of 20 mothers (10 intervention and 10 controls) in order to coordinate food sampling at the same time between an intervention household and its control counterpart.

Field workers trained the mothers of the intervention group during three weeks, assessing their knowledge, observing them during the whole process, noting deviations and recommending corrective measures.

After three weeks training, food samples were collected in both intervention and control households and examined in the National Laboratory of Public Health.

Three months later, a third round of food sample collection and examination was conducted to assess the persistence of behaviour change in the Intervention Group. At this stage 15 intervention mothers were observed during foods preparation and 15 were not observed, in order to assess the influence of the field workers' presence on the mothers' behaviour.

FC are indicators of a serious food borne hazard, and are extremely heat sensitive (NSF, 2006). FC are also the main indicator used to monitor microbiological quality of water and food in Mali (DNS, 2004). Indeed, they are the sole faecal strains that the National Laboratory of Public Health is able to identify and count. Therefore we considered FC as the sole indicator of the safety of our intervention food samples. Laboratory analyses were directed at identification and counting of FC colonies.

FC counts from the Intervention Group before and after the intervention were compared to assess the effectiveness of the intervention in reducing FC organisms in weaning foods. The Control Group FC count was used to follow bacteriological contamination trend of foods over the seasons. We concentrated on sampling foods for FC count at the moment of feeding children with cooled foods, after cooking and reheating.

The experimental study demonstrated that cooking was effective in FC elimination; indeed no FC were found in food samples during the experiment. In addition no difference was found between cooking and reheating temperatures ($P < 0.26$ and 0.071 respectively for Moni and Fish Soup, section 2.11, Table 2.16). These findings were confirmed by the baseline measured cooking temperatures (at least 97.7°C and 97.6°C for Moni and Fish Soup respectively). However both cooked and reheated foods were found contaminated after some minutes of cooling, indicating that cross contamination occurred due to mothers' unhygienic foods handling at this step. Then food samples were taken only at the two remaining CCP (child feeding with cooled foods after cooking and after reheating). In addition, this decision was based upon the argument that to cause illness an infectious dose of bacteria must be ingested, (Keusch, 2006; Lanata, 2003). Then the most relevant CCPs were cooled foods ready for feeding children, following cooking and reheating. Therefore we concentrated sampling for FC count at the moment of child feeding with cooled foods, after cooking or reheating, during the intervention and its assessment. Particular attention was given to hygiene messages (washing hands with clean water and soap at critical periods, washing dishes with clean water and soap...).

Cooking and reheating to temperatures above 70°C during at least 10 minutes were selected as monitoring criteria. It was found during the experimental phase of the study (Chapter 2) that mothers' traditional cooking habits were effective to eliminate FC, and reheating (if adopted) was as effective as cooking. Criteria were therefore monitored only to confirm these experimental findings.

No bacteriological standards are available for the two local meals considered, thus, taking into account the hazardous environment in which the intervention occurred, and the capacity of FC detection of our enumeration method, we decided that 10 FC/g was a reasonable target. This threshold (10 FC/g) was selected to assess the performance of the intervention.

3.2. Specific objectives

The two specific objectives of this phase were:

- To eliminate or reduce to an acceptable level, at CCPs, FC contamination in weaning foods through messages delivered at household level;

- To develop recommendations on how to apply the HACCP approach to domestic food preparation in developing countries.

3.3. Recall of educational messages

The messages selected to induce implementation of corrective actions (see lessons learned in experimental Phase, Chapter 2) were:

- Reheating meals at boiling point even if only for a few seconds (The state of boiling indicated by the appearance of steam, is easier to monitor by mothers who have no thermometer);
- Using clean water (from a tap or borehole) for home needs (cooking, drinking, washing dishes,); When washing dishes, running water and soap must be used;
- Washing hands with running clean water and soap at critical periods: these critical periods are: after using latrines or cleaning a child's bottom, before handling food, and before feeding a child.

3.4 Participant sample size calculation

The objective is to demonstrate that the intervention improves foods' bacteriological quality for a proportion of 70% more than no intervention, with 90% power of achieving a significant result at the 5% level.

We used the general formula (Kirkwood, 2003)

$$n \geq \frac{\left[\frac{u \sqrt{\pi(1-\pi)}}{(\pi - \pi_{null})^2} \left(+v \sqrt{(\pi_{null}(1-\pi_{null}))^2} \right) \right]}{(\pi - \pi_{null})^2}$$

n , required minimum sample size

$\pi = 0.7$, proportion of interest

$u = 1.28$, one-side percentage point of the normal distribution corresponding to 10%, if the power is 90%

$v = 1.96$

$\pi_{null} = 0.5$, nul hypothesis proportion

$n \geq 60$, then we considered 60 mothers as our study sample.

3.5. Selection of Participants

To get the new sample for this phase of the study, we visited the nine Community Health Centres of Commune V. At the time of the nutritional education sessions during each visit, we listed all mothers present of children aged from six to nine months. For each mother, the name, age and sex of her child were mentioned as well as her name, home address and telephone number (if available). Health Centre visits continued until we had 120 volunteer mothers.

From the list of 120 mothers, 60 mothers were selected randomly and split in two groups of 30. Visits of selected mothers at home were undertaken to locate their household, and to confirm their agreement to be involved in the study. Two selected mothers who reconsidered their participation were replaced by two others selected from the rest of the list of 120 mothers.

During the recruitment of mothers we emphasized our need to be able to visit their household for observation of weaning foods preparation, handling and child feeding at any time in the day, and to take food samples, and obtained their consent.

Our team of field workers identified households where mothers included in our sample live. During this first visit we obtained authorization to visit households at any time in accordance with the study plan. Household characteristics forms were also completed.

3.6. Intervention approach

3.6.1. Onset

The first mothers' household visit, after its location identification, was focused on an assessment of their actual practices regarding corrective actions selected for the intervention, through one day observation. Of concern were that mothers may not reveal their true practices, if they know the purpose of our intervention (Curtis et al, 1998; Monte et al, 1997). So, at this stage, mothers were informed that our purpose was neither to create a competition between them nor to evaluate their capabilities, but to learn about their weaning food recipes, that could help formulate new weaning foods to tackle malnutrition.

During this first visit, dedicated also to building confidence among fieldworkers and mothers and to enhancing their self-esteem, it was indicated that any suggestion could be very useful for the following steps of the intervention.

With the selected food planned to be prepared that morning, the fieldworker started observation of the whole process of preparation, handling, storage and child feeding. The purpose of these observations was to identify mothers' actual practices regarding the compliance with corrective actions, and so to improve if necessary the content of messages in order to meet the precise need of each mother.

As a compensation for demonstrations of meal preparation and sampling of foods, an amount of 2000 CFA (local currency), approximately £2, was allocated on the eve of each demonstrative session to each mother.

Sampling of weaning foods in the 60 volunteers' households for FC count in the Laboratory of Public Health, was conducted as a baseline of the intervention.

At the end of this step, the 60 mothers were split in two groups of 30 mothers each. The first group, which was intended to receive intervention messages, was termed the "Intervention Group", and mothers involved designated as intervention mothers. The second group of 30 (not receiving messages) was considered as the "Control Group", and members termed control mothers.

Only the two kinds of foods (Moni and Fish Soup examined during the HACCP experiment and for which CCP were well established) were targeted in this intervention.

3.6.2. Intervention group training and observation

This group was split again into three sub-groups of ten mothers. Each sub-group were followed by a field worker. This latter's duties were household visits to train and observe mothers. Visits started at around 9 am and lasted one to two hours in the morning, and at around 3 pm and lasted one hour in the afternoon. Each intervention mother was visited twice a day every two weeks to:

- deliver messages (explanation of what is considered as a clean water, where to get it and how to store it in a safer manner; description of boiling status) and guidance during training sessions on how to perform corrective actions (demonstration of washing hands and utensils with clean water and soap),
- monitor the Intervention Group, and help the laboratory staff with foods sampling,

These fortnightly visits continued for 9 months.

The second visit to each intervention mother, after the identification of households' location was focused on:

- clarification of the actual objectives of the intervention,
- delivering messages,
- Demonstrations, e.g. of hand washing practices,
- Recall of corrective actions, taking into account deviations observed during observations.

Mothers were always asked to notify constraints they may have had in implementing corrective actions effectively, and to suggest possible solutions. For instance, some mothers said that they could not afford to buy soap regularly. Thus, Hand washing kit (equipment and soap) (Figure 2.13, section 2.8, Chapter 2) was provided to all of them by the fieldworker during the intervention period.

In households where other family members were involved in the child's care, they were also included in message delivery and followed for compliance with corrective actions. In this case, the intervention mother was strongly advised to support her child's care giver to follow the field worker's recommendations.

Field workers were provided with a form to record relevant information related to mothers' compliance with corrective actions, and corrections undertaken.

The field worker wrote down all deviations from corrective actions that occurred during a daily observation of the mother's performance in weaning food preparation and handling. At the end of each session, the fieldworker discussed with the mother any non-performance observed (deviations from corrective actions). Field workers noted explanations given by mothers on the non compliance with corrective actions and their suggestions if any. Field workers recalled corrective messages and requested an appointment for the following week. This latter session began with a recall of the previous week's deviations observed and the corrective actions needed for their compliance.

The training lasted three weeks at the end of which mothers underwent an assessment. After these weeks' training, a period for message enhancement and support (Monte, 1997), food sampling started. Physical parameters (temperatures of foods, ambient air and in storage room, humidity and pH) were recorded on-site using Klipspringer Instrument equipment as in the experimental phase.

3.6.3. Control Group follow-up

The control group, that did not receive any messages or training, were also well identified by field workers in order to assist the laboratory experts in food sampling. Food samples were taken for FC unit count at the time when the intervention household's foods were sampled. Indeed field workers managed to schedule each intervention mother with her counterpart control mother. Information about the study's true objectives was not given to control mothers so that they continued to prepare and handle their children's weaning foods as usual. However they benefited from allocations given to the intervention group: a hand washing kit and the amount of £2 on the eve each food preparation session.

3.6.4. Evaluation of the intervention and assessment of its sustainability

An evaluation at the end of the intervention and another assessment three months later were carried out to find out: firstly the effectiveness of the intervention in improving the bacteriological safety of foods prepared by mothers who were not under direction (though they might be under observation), and secondly how long the changed behaviour could last and, what could be its impact on performance of intervention mothers in the longer term.

Given that this group was observed during all previous steps, half of them were not observed when preparing and handling their foods so that we could find out the impact of observations on mothers behaviours. Bacteriological quality (FC count) of foods prepared by 15 observed mothers and 15 non- observed mothers were compared.

3.7. Food sampling

During the intervention period (nine months), food samples were taken in each of the 60 households at the moment of child feeding with foods cooled after cooking or reheating. Food sampling happened three times following local seasons (hot season: June, rainy season: August and cold season: December), in order to take into account seasonal variations (Henry et al, 1990, Lanata et al, 2003). Sampling time were coordinated between field workers and laboratory staff, the former liaising with the households and helping the latter to know the right moment in the morning and in the afternoon for food sampling. 720 food samples were taken and analysed at the National Laboratory of Health for FC counting in the same conditions, with the same equipment as in the experimental phase (Chapter 2). It is important to notice that the Laboratory technicians were blinded to the Intervention and Control Groups.

3.8. Data analysis

EpiDATA analysis 1.1 was used to describe physical parameters; means, maxima and minima were calculated for 95 % confidence interval (95% CI).

FC colony counts were transformed logarithmically to calculate geometric means. Microsoft Excel 2007 Student's t-test, two-tailed type 3 was used to compare means of all parameters, considering a difference significant if $P < 0.05$; 95 % Confidence Intervals were also calculated.

The intervention's performance (RR and P Value) in reducing FC contamination at CCP was calculated using Epi-info 6.4 KH2 considering each difference significant if $P < 0.05$; the 95 % confidence interval (CI) was also calculated for each food.

3.9. Ethical clearance:

To undertake data collection, local authorities and community health centres' Chairpersons and directors signed a memorandum expressing their commitment to support the study team on March 25th 2007. Clearance was obtained from the Malian national ethics and life sciences committee on April 13th 2007 and the LSHTM ethics committee on September 20th 2007. A consent form showing agreement for involvement in the study and authorization for pictures taken for the study was signed by each study sample mother or her husband. An amount of about £2 was given to mothers as a compensation for purchase of ingredients; a hand washing kit (equipment and soap) was also distributed to mothers of both intervention and control groups. For the latter, the kit was given at the end of the study so as to avoid changing their food preparation and handling habits.

3.10. Results

3.10.1. Demographic and socio-economic profile of the study sample

Table 3.1: Characteristics of children (N=60)

Parameters	Number reported (%)		
	Intervention group	Control group	Total
Age (months)			
6	6(20.0)	5(16.7)	11(18.3)
7	5(16.7)	4(13.3)	9(15.0)
8	2(6.7)	3(10.0)	5(8.3)
9	17(56.7)	18(60.0)	35(58.3)
Sex			
Male	5(15.0)	12(40.0)	27(45.0)
Female	15(50.0)	18(60.0)	33(55.0)

Table 3. 2: Parents' characteristics

Parameter	Number reported (%)					
	Intervention group		Control group		Total	
	Mothers	Fathers	Mothers	Fathers	Mothers	Fathers
AGE						
15-24	12(40.0)	1(3.3)	9(30.0)	1(3.3)	21(35.0)	2(3.3)
24-34	15(50.0)	15(50.0)	16(53.3)	7(23.3)	31(51.7)	22(36.7)
35+	3(10.0)	14(46.7)	5(16.7)	22(73.4)	8(13.3)	36(60)
TOTAL	30(100)	30(100)	30(100)	30(100)	60(100)	60(100)
EDUCATION						
No schooling	15(50.0)	10(33.4)	19(63.3)	13(43.3)	34(56.7)	23(38.3)
P. school	10(33.3)	6(20.0)	9(30.0)	13(43.3)	19(31.6)	19(31.6)
S. school	4(13.3)	7(23.3)	2(6.7)	3(10.0)	6(10.0)	10(16.7)
P. S. school	1(3.4)	7(23.3)	0.0	1(3.4)	1(1.7)	8(13.4)
TOTAL	30(100)	30(100)	30(100)	30(100)	60(100)	60(100)
OCCUPATION						
House wife	14(46.7)	0.0	15(50.0)	0.0	29(48.3)	0.0
Farmer	0.0	0.0	0.0	1(3.3)	0.0	1(1.7)
Seller	3(10.0)	0.0	4(13.3)	0.0	7(11.7)	0.0
Artisan	5(16.7)	2(6.7)	6(20.0)	6(20.0)	11(18.3)	8(13.3)
Worker	0.0	3(10.0)	0.0	3(10.0)	0.0	6(10.0)
Driver	0.0	2(6.7)	0.0	2(6.7)	0.0	4(6.7)
Trader	4(13.3)	11(36.7)	4(13.3)	12(40.0)	8(13.3)	23(38.3)
Entrepreneur	0.0	1(3.3)	0.0	1(3.3)	0.0	2(3.3)
Civil servant	0.0	8(26.6)	1(3.4)	2(6.7)	1(1.7)	10(16.7)
Other	4(13.3)	3(10.0)	0.0	3(10.0)	4(6.7)	6(10.0)
TOTAL	30(100)	30(100)	30(100)	30(100)	60(100)	60(100)

All of the study children were aged between 6 and 9 months. The latter was the most important age group in both Intervention and Control Group. While male and female are equal in the Intervention Group, females represent 60% of the Control Group. The age interval (6-9 months) was preferred, because at the end of the nine months period of our study the oldest children were 18 months old. Above this age many mothers stop using weaning foods; consequently they could withdraw from the sample before the end of the study.

Mothers were very young in the two groups, 52% of them were under 35. Fathers were older, only 22% of them are under 35.

Literacy level was very low in both sexes, although men were more educated. Only one mother out of 60 was a civil servant. About 50% of mothers were illiterate and were housewives (have no independent income).

Fathers were older than mothers, about 50 % of the latter were aged less than 34 years, whereas more than 50% of the former were aged more than 35 year .

The literacy level was low in both Mothers and Fathers groups, whilst the latter were more educated .

About 50 % of Mothers were housewives without independent income; however some of them were active out of their household. All Fathers are active even though most of their occupations are very low-paid. These households' status is consistent

with the experimental phase (Chapter 2) in that they mostly all pertained to the low income class.

3.10.2. Bacteriological results

3.10.2.1. Characteristics of baseline food samples' physical parameters at CCPs

A) Description of physical parameters

Table 3.3: Description of baseline physical parameters of samples measured at CCP (N= 60)

Variable	Mean	Sdev	Max	Min
Moni				
Cooking T (°C)	98.69	0.59	102.00	97.90
Cooking duration (min)	27.08	16.40	75.00	4.00
Cooled food T(°C)	39.50	2.60	48.30	30.00
Cooked food pH	4.71	0.41	6.55	2.74
Leftover storage duration (min)	283.35	43.15	402	191.00
Leftover T(°C)	35.99	2.56	46.10	28.40
Leftover pH	4.57	8.72	6.59	2.01

Fish Soup				
Cooking T (°C)	99.00	0.43	100.00	97.60
Cooking duration (min)	28.88	19.33	87.00	7.00
Cooled food T(°C)	37.69	3.28	47.60	30.00
Cooked food pH	5.62	0.42	6.60	4.85
Leftover storage duration (min)	267.27	40.22	362.00	173.00
Leftover T(°C)	34.19	2.20	41.20	30.08
Leftover pH	5.68	0.43	6.65	4.30

N – number of measurements for each parameter

Table 3.3 indicates that cooking temperatures of both Moni and Fish Soup at all CCP ranged between 97°C and 102°C. Cooking durations of the two foods also ranged between 4 and 87 min. In general the pair of parameters time and temperature of cooking was far above the values required for pathogen destruction (60°C, 12 min) (Bryan et al, 1992); even for the few samples for which cooking duration was less than 12 min, this deviation was compensated by the very high temperature. These high values of these two parameters resulted in the complete elimination of FC in foods observed over the experiment.

Food temperatures at the moment of feeding children ranged between 30 and 48°C for both Moni and Fish Soup after cooking and after storage (leftover). These temperatures were comparable to ambient indoor temperatures in the study area and to those suitable for pathogen growth (Bryan et al, 1992; Banward et al, 1995).

Leftover storage durations ranged between 173 min and 402 min, sometimes more than 6 hours and always more than 2 hours. These very favourable conditions for pathogen growth resulted in a high contamination of both Moni and Fish Soup observed during the experimental phase.

The pH of food ranged between 2.01 and 6.65, the values less than 7 meaning that foods were acid or slightly acid, due probably to the use of ingredients like monkey bread (baobab fruit) or lemon in Moni, and lemon and vegetables in the Fish Soup.

B) Compararison of physical parameters of two weaning foods

Table 3.4: Compararison of physical parameters of two weaning foods: P values relative to the null hypothesis of no difference between Intervention and Control Groups at baseline. N = 60

Variable	P (Student's t test)
Moni	
Cooking Temperature (°C)	< 0.230
Cooking duration (min)	< 0.601
Cooled food Temperature(°C)	< 0.730
Cooked food pH	< 0.760
Leftover storage duration (min)	<0.610
Leftover Temperature (°C)	<0.580
Leftover pH	<0.340
Fish Soup	

Cooking Temperature (°C)	<0.230
Cooking duration (min)	<0.601
Cooled food Temperature (°C)	<0.730
Cooked food pH	<0.761
Leftover storage duration (min)	<0.061
Leftover Temperature (°C)	<0.587
Leftover pH	<0.337

The data in Table 3. 4 above indicate that there was no significant difference between Intervention and Control Group regarding baseline physical parameters.

3.10.2.2. Bacteriological parameters

A) Study Control Group results and analysis

Sampling of foods was carried out over the three local seasons: dry season (April-June), rainy season (July- September) and cold season (December- February). FC count associated to seasonal variation was determined. Were also calculated FC count differences between groups at each CCP.

• Results

Table 3, 5: FC count distribution (FC/g) over seasons in Moni Control Group

Range (CF/g)	June		August		December	
	After cooking	After storage	After cooking	After storage	After cooking	After storage
	No. of samples (% of column total)					
ND	9 (30.0)	0 (0.0)	1 (3.3)	1 (3.3)	1(3.3)	0 (0.00)
10-100	5 (16.7)	1 (3.3)	2 (6.7)	0(0.0)	4 (13.3)	1(3.3)
101-1000	16 (53.3)	25 (83.3)	25 (83.3)	25 (83.3)	25 (83.3)	27 (90.0)
1001- 10 ⁴	0 (0.0)	4 (13.3)	2 (6.7)	4(13.3)	0 (0.0)	2 (6.70)

N	30 (100)	30(100)	30(100)	30(100)	30(100)	30(100)
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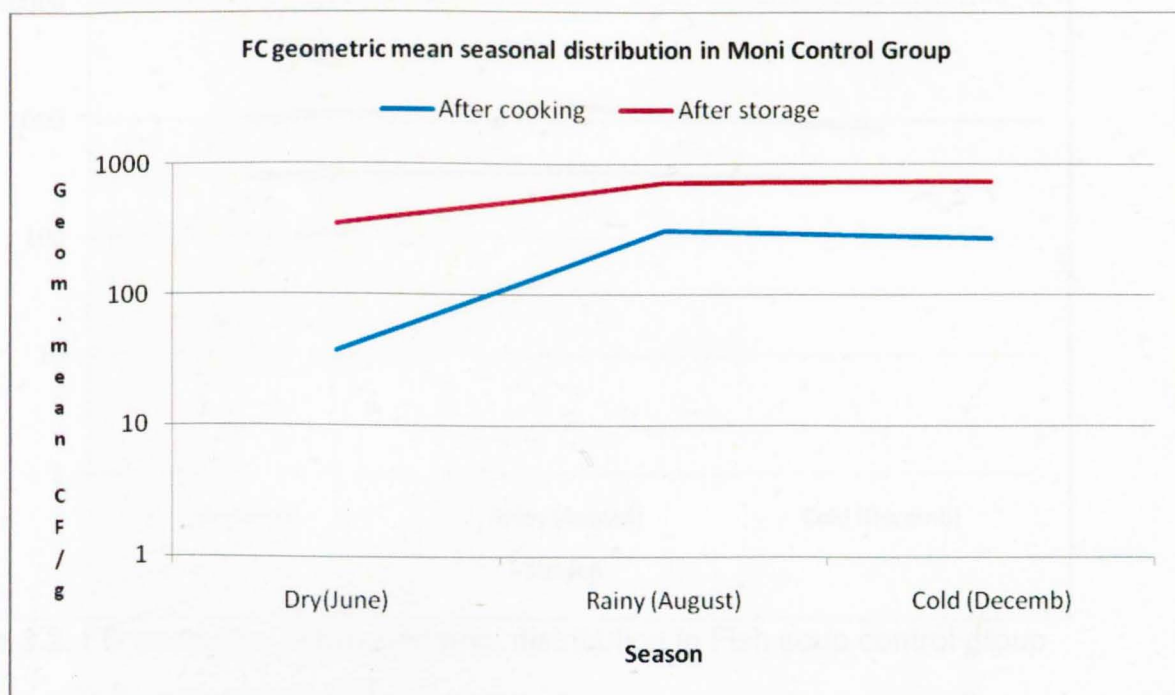


Figure 3.1: FC geometric mean seasonal distribution in Moni Control Group

Table 3.6: FC count distribution (FC/g) over seasons in Fish Soup Control Group

Range (FC/g)	June		August		December	
	After cooking	After storage	After cooking	After storage	After cooking	After storage
	No. of samples (% of column total)					
ND	0(0.0)	0(0.0)	3 (10.0)	0(0.0)	1 (3.3)	0 (0.0)
10-100	1(3.3)	1(3.3)	0(0.0)	1(3.3)	3 (10.0)	1 (3.3)
101-1000	28 (93.3)	19(63.3)	27 (90.0)	9(230.0)	26 (86.7)	28(93.4)
1001-10 ⁴	1 (3.3)	10 (33.4)	0(0.0)	20 (66.7)	0(0.0)	1(3.3)
N	30 (100)	30 (100)	30 (100)	30 (100)	30 (100)	30 (100)

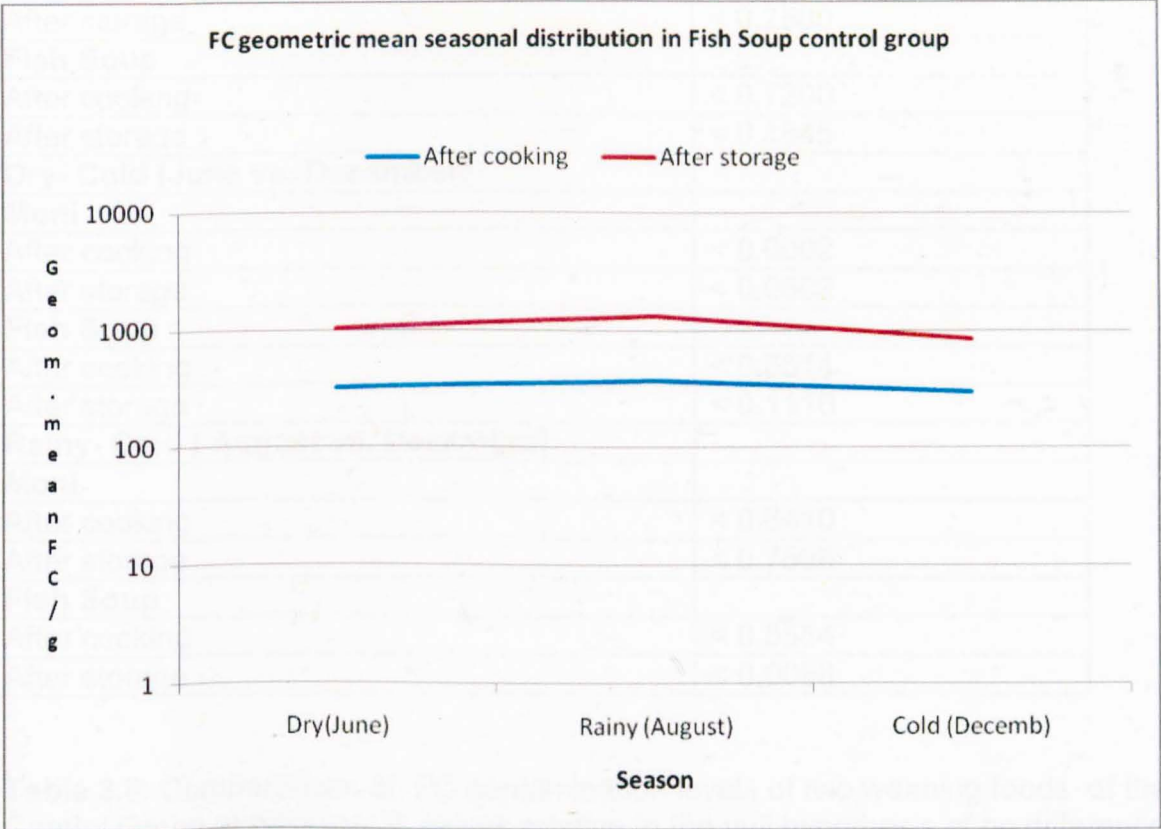


Figure 3.2: FC geometric mean seasonal distribution in Fish soup control group

The contamination of most of samples in Moni (from 53% to 90%) ranged between 100 and 1000 FC /g ; More than 96% of all samples, except those of June after cooking, contamination level was higher than 100 FC/g. 30% of June after cooking samples contained less than 10 FC/g, whilst in December and in August, contamination in all stored food samples was above this level (Table 3.5).

For both processes, after cooking and after storage, the contamination in Moni was higher in rainy and cold seasons than in dry season (Figure 3.1). This Figure shows also that stored foods were always more contaminated than after cooking foods.

Fish Soup appeared to be more likely exposed to high FC contamination and less subject to seasonal variation (Table 3.6 and Figure 3.2). As in Moni, Fish Soup stored samples were always more contaminated than after cooking ones. In addition stored samples appeared to be more contaminated in rainy season than in cold and dry seasons, whilst after cooking samples contamination trend seemed to be similar in all seasons.

• **Samples Analysis**

Table 3.7: Comparison of FC contamination levels of two weaning foods in the Control Group; P values relative to the null hypothesis of no difference between seasons

SEASONS	P (Student's t test)
Dry- Rainy (June vs. August)	
Moni	
After cooking	< 0.0004

After storage	< 0.2860
Fish Soup	
After cooking	< 0.7200
After storage	< 0.1845
Dry- Cold (June vs. December)	
Moni	
After cooking	< 0.0002
After storage	< 0.0602
Fish Soup	
After cooking	< 0.6514
After storage	< 0.1116
Rainy- Cold (August vs. December)	
Moni	
After cooking	< 0.6410
After storage	< 0.7598
Fish Soup	
After cooking	< 0.5584
After storage	< 0.0098

Table 3.8: Compararison of FC contamination levels of two weaning foods of the Control Group at baseline; P values relative to the null hypothesis of no difference between two steps (after cooking and after storage) for each food

Food	Step	FC/g Geometric mean	P
Moni:	After cooking	37	< 0.0001
	After storage	309	
Fish Soup:	After cooking	316	< 0.0001
	After storage	708	

The data in Table 3.7 above indicate a significant difference in Moni contamination levels after cooking between Dry and Rainy season and between Dry and Cold season. There was also a significant difference in Fish Soup contamination after storage between Rainy and Cold seasons. These results were consistent with the FC contamination trend shown in Figures 3.1 and 3.2. Indeed after cooking, Moni samples were more contaminated in August and December than in June, as were Fish soup foods after storage in December compared to those of August and June.

There was a significant difference in FC contamination levels in both Moni and Fish Soup, between the two steps. Stored foods were more contaminated than foods cooled after cooking (Table 3.8). Indeed leftover foods were stored at least two hours at room temperature before service.

B) Intervention Group results

- Intervention group mothers' training

To assess trainees it was considered that a message was well understood when the mother implemented accurately the corresponding corrective action to control FC at CCP at anytime. For example, the following were considered inaccurate implementation: washing one hand or washing hands without soap or forgetting to wash hands at critical moments, during the process of food preparation and handling. A mother was asked to perform preparation and handling of one of the two foods selected. The field worker observed and took note of all deviations seen, but was instructed to make no comment. Table 3.9 below displays the number and the corresponding percentage of mothers who performed accurately each message considered. This assessment was undertaken prior to sampling the food, in order to evaluate the effectiveness of the CCP intervention in controlling FC contamination.

Table 3.9: Messages understood and seen to be implemented, after three weeks of training (30 intervention group mothers)

Behaviour Period	No. of mothers observed performing the correct behaviour (%)			
	Potable water usage (%)	Washing dishes (%)	Hand washing (%)	Reheating leftovers (%)
End of training	27 (90.0)	26 (86.7)	21 (70.0)	30 (100)
Moni sampling	30 (100)	30 (100)	30 (100)	30 (100)
Fish Soup sampling	30 (100)	30 (100)	29 (96.7)	30 (100)
Remarks	Only water from tap or bore hole	Local soap included	Deviation when one critical step missed	Sometime after appearance of vapour

Table: 3. 9 indicates that most of the mothers mastered the corrective actions, mainly the reheating of leftovers, after three weeks training. However, hand washing scores remained less than the other measures. Surprisingly, reheating (which is not a local habit) was completely adopted by mothers.

Finally, their performance during food preparation confirmed the mothers' good understanding of the messages.

- **Intervention Group mothers ability to control FC contamination of foods at CCPs evaluation**

Many social studies have shown that there were disparities between individuals' knowledge of specific hygiene behaviours and their implementation of these behaviours. This step aimed to assess how effective was their understanding of the messages on mothers' ability to control FC contamination at CCPs of the two foods selected. Food samples were taken when mothers were performing food preparation and handling, and analysed by an independent specialist of the National Laboratory of Public Health for FC count.

Table 3.10: Moni Intervention Group before intervention (baseline) FC count (FC/g) at CCPs

Range (FC/g)	No. of samples (% of column total)			
	Intervention (baseline)		Control	
	After cooking	After storage	After cooking	After storage
ND	17 (56.7)	1 (3.3)	9 (30.0)	0 (0.0)
10-100	4 (13.3)	9 (30.0)	5 (16.7)	1 (3.3)
101-1000	9 (30.0)	18 (60.0)	16 (53.3)	25 (83.3)
1001-10 ⁴	0 (0)	2 (66.7)	0 (0.0)	4 (13.3)
N	30(100)	30(100)	30(100)	30(100)

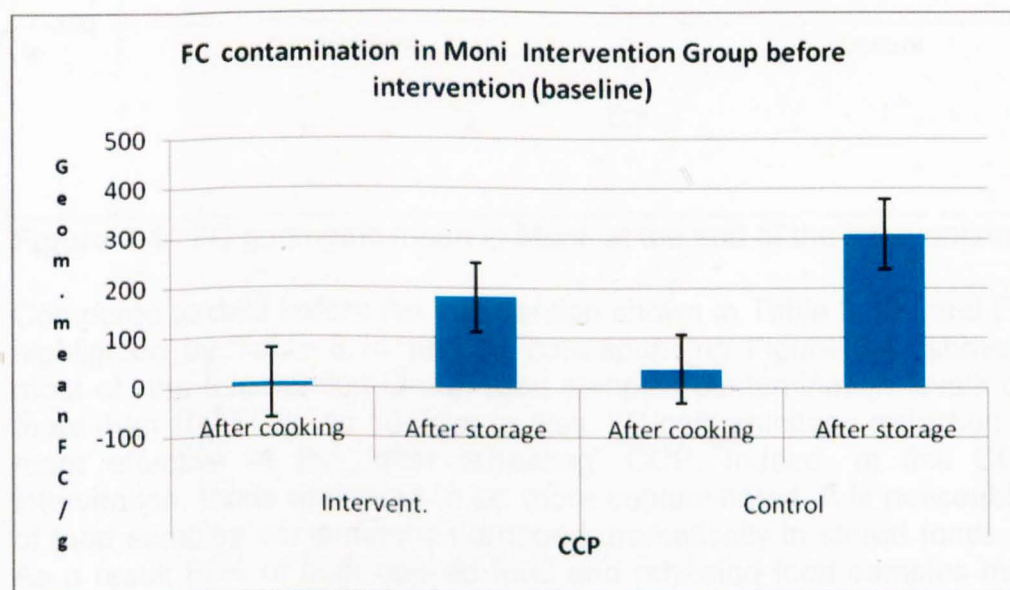
**Figure 3.3:** FC geometric means in Moni before intervention (baseline)

Table 3.10 shows that most of the Moni samples' contamination levels ranged between 10 and 1000 FC/g. However, more than 56 % of the samples of foods after cooking, from the Intervention Group, counted less than 10 FC/g. In both Intervention and Control Group, after storage samples seemed to be more contaminated than after cooking ones (Figure 3.3).

Table 3.11: Moni Intervention Group FC count (FC/g) at the end of the intervention at CCPs

Range(FC/g)	No. of samples (% of column total)			
	Intervention		Control	
	After cooking	After reheating	After cooking	After storage
ND	25 (83.3)	25 (83.3)	1 (3.3)	1 (3.3)
10-100	2 (6.7)	4 (13.3)	2 (6.7)	0(0.0)
101-1000	3 (10)	0 (0.0)	25 (83.3)	25 (83.3)
1001-10 ⁴	0 (00)	1 (3.3)	2 (6.7)	4(13.3)
N	30 (100)	30 (100)	30 (100)	30 (100)

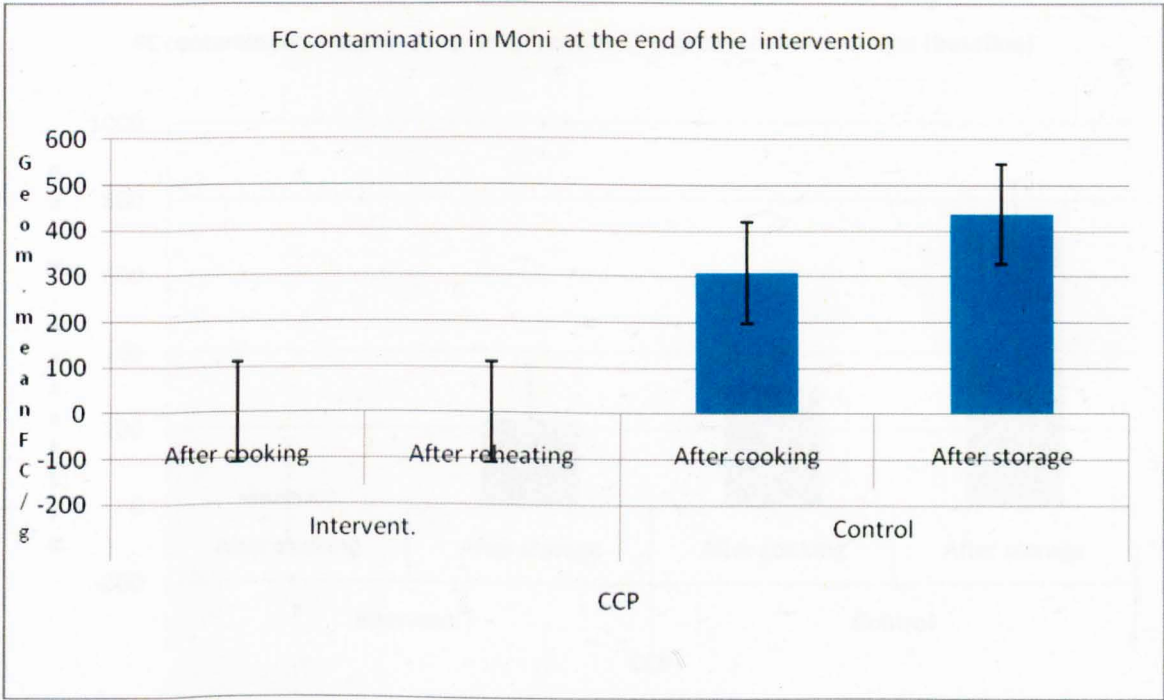


Figure 3.4: FC geometric mean in Moni at the end of the intervention

Compared to data before the intervention shown in Table 3.10 and Figure 3.3, data highlighted by Table 3.11 and its corresponding Figure 3.4 (above), indicate that most of the Intervention Group food samples contamination levels decreased from more than 100 FC/g to 10 FC/g or less. FC contamination reduction appeared to be more effective at the “after reheating” CCP. Indeed, at this CCP, before the intervention, foods appeared to be more contaminated. It is noticeable that the level of food samples contamination dropped dramatically in stored foods after reheating. As a result 83% of both cooked food and reheated food samples met the standard that was set (<10FC/g).

In the Control Group the contamination trend was similar to that observed before the intervention. The contamination level of most samples (more than 90%) ranged between 10² and 10⁴ FC/g, and many counted more than 10³ FC/g. Control Group stored food samples appeared to be the most contaminated (Figure 3.3).

Table 3.12: Fish Soup Intervention Group before intervention (Baseline) FC count (FC/g) at CCPs

Range(FC/g)	No. of samples (% of column total)			
	Intervention (baseline)		Control	
	After cooking	After storage	After cooking	After storage
ND	9 (30.0)	2 (6.7)	0(0.0)	0(0.0)
10-100	9 (30.0)	3 (10.0)	1(3.3)	1(3.3)
101-1000	12 (40.0)	22 (73.3)	28 (93.3)	19(63.3)
1001-10 ⁴	0 (00)	3 (10.0)	1 (3.3)	10 (33.4)
N	30 (100)	30 (100)	30 (100)	30 (100)

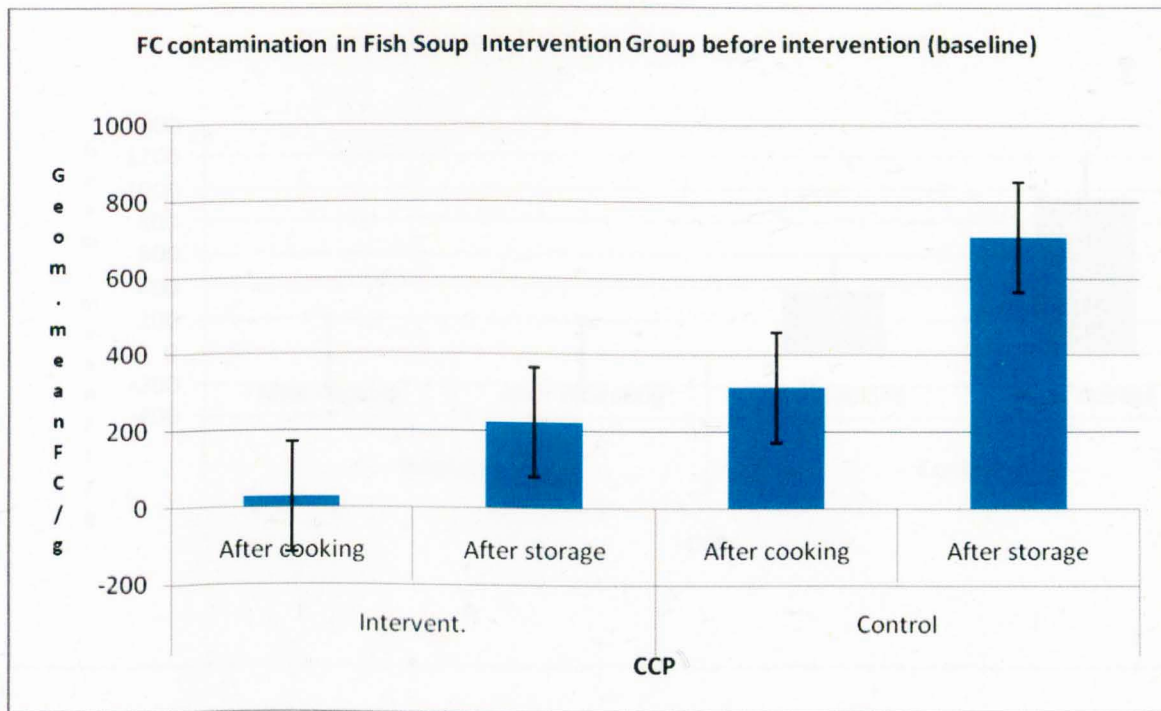


Figure 3.5: FC geometric mean in Fish soup before intervention (baseline)

Table 3.12 data show that most of Fish Soup samples' contamination levels ranged between 100 and 1000 FC/g. It is noticeable that on one hand, all the Control Group samples contained more than 10 FC/g, and on the other hand more than 93 % of them contained more than 100 FC. The highest level of contamination appeared in stored food samples into the two groups. About 30% and 6.7% of the intervention group after cooking and after storage, respectively, already met the standard set (10 FC/g).

Table 3.13: Fish Soup Intervention Group FC count (FC/g) at the end of the intervention at CCPs

Range(FC/g)	No. of samples (% of column total)			
	Intervention		Control	
	After cooking	After reheating	After cooking	After storage
ND	26 (86.7)	29 (96.7)	3 (10.0)	0(0.0)
10-100	4 (13.3)	0 (00)	0(0.0)	1(3.3)
101-1000	0(00)	0 (00)	27 (90.0)	9(230.0)
1001-10 ⁴	0(00)	1 (3.3)	0(0.0)	20 (66.7)
N	30 (100)	30 (100)	30 (100)	30 (100)

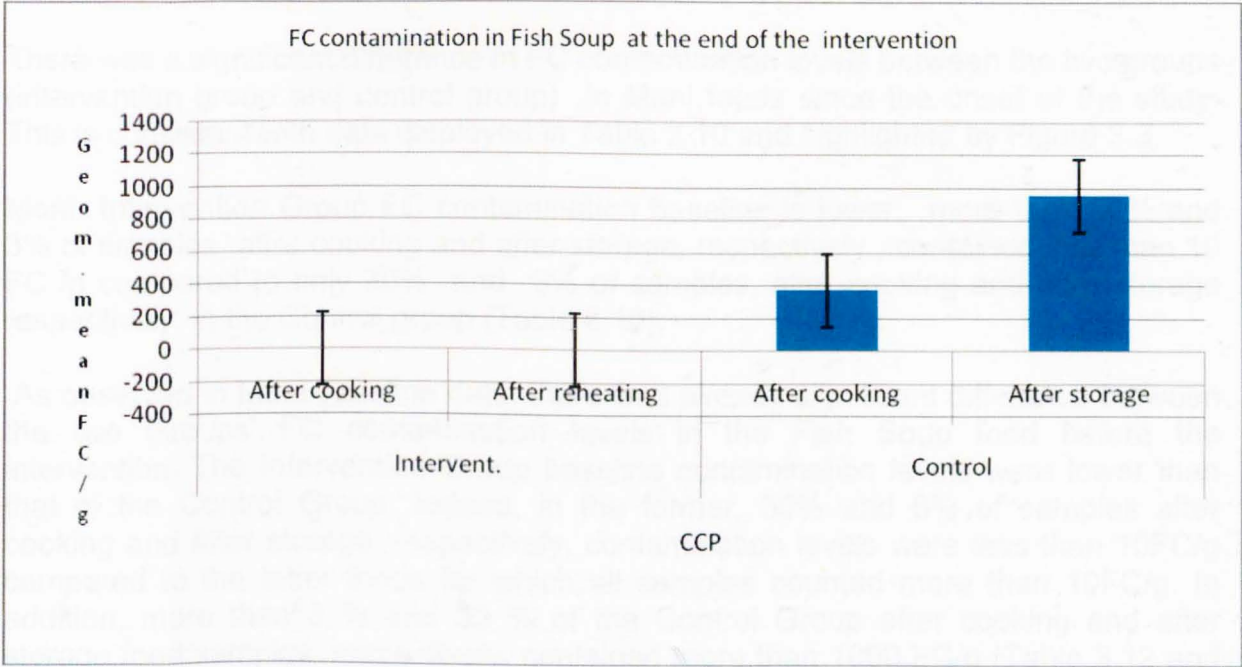


Figure 3.6: FC geometric mean in Fish Soup at the end of the intervention

Compared to Table 3.12 data and its corresponding distribution in Figure 3.5, Table 3.13 data highlighted by Figure 3.6 indicate a dramatic drop of the contamination level at CCPs in the Intervention Group both after cooking and after reheating. About 87% and 97% of after cooking and after reheating samples, respectively met the standard set (10FC/g)

The Control Group contamination trend was similar to that observed before the intervention for both Moni and Fish Soup after cooking and after reheating. However, stored food samples appeared to be the most contaminated, about 50% of them contained more than 1000 FC/g.

• **Data analysis**

Table 3.14: Compararison of FC contamination levels of two weaning foods P values relative to the null hypothesis of no difference between Intervention and Control Group before the intervention (Homogeneity test)

CCP	P
Moni	
After cooking	< 0.0200
After storage	< 0.008
Fish soup	
After cooking	< 0.0001
After storage	< 0.002

There was a significant difference in FC contamination levels between the two groups (intervention group and control group) in Moni foods since the onset of the study. This is consistent with data displayed in Table 3.10 and highlighted by Figure 3.3.

Moni Intervention Group FC contamination baseline is lower; more than 56% and 3% of samples after cooking and after storage, respectively, contained less than 10 FC /g compared to only 30% and 0% of samples, after cooking and after storage respectively, in the Control group (Table 3,10).

As observed in Moni baseline data, there was already significant difference between the two Groups' FC contamination levels in the Fish Soup food before the intervention. The Intervention Group baseline contamination levels were lower than that of the Control Group. Indeed, in the former, 30% and 6% of samples after cooking and after storage, respectively, contamination levels were less than 10FC/g compared to the latter foods for which all samples counted more than 10FC/g. In addition, more than 3 % and 33 % of the Control Group after cooking and after storage food samples, respectively, contained more than 1000 FC/g (Table 3.12 and Figure 3.5).

According to these results, the two groups (Intervention and Control) are not comparable). However, the Control Group FC contamination trend over the seasons indicated that, without the intervention, the FC contamination levels in the Intervention Group will be similar at anytime in the year. Indeed, whilst the FC contamination levels decreased dramatically in the Intervention Group foods after the intervention, the FC contamination trend of foods in the Control Group remained constant over the study period. On the other hand, the effectiveness of the intervention was assessed by comparing the FC contamination levels of foods of the Intervention Group before and after the intervention.

• **Intervention performance**

How effective was the intervention in preventing foods FC contamination? The size of this effect is measured by the Relative Risk (RR), the ratio of the risk of FC contamination in the Intervention Group with or without intervention. The higher is RR the more effective is the intervention in FC reduction in foods.

The main objective of food safety management is to reduce hazards to levels at which consumers' health is no longer at risk. The criteria to assess these levels of hazards reduction are standards. As there were no official standards for these local foods, it is considered in this study that FC contamination reduction to less than 10

FC/g is a good performance. Therefore this threshold was considered to assess the performance of the HACCP approach.

Table 3.15: Association between the intervention and FC contamination reduction at CCPs in two foods

CCP	RR	95%CI
Moni after cooking	5.80	2.60-12.95
Moni after reheating	4.83	2.36-9.92
Fish soup after cooking	6.77	2.69-16.94
Fish soup after reheating	15	3.93-57.22

These results highlight the effectiveness of the HACCP approach in foods microbiological safety improvement, but it appeared that the intervention was more effective in controlling FC contamination in Fish Soup food at both CCPs (after cooking and after reheating) with respectively RR = 6.77 after cooking and RR= 15 after reheating.

Surprisingly, in Fish Soup, reheating appeared to be more protective. Indeed, more than 96% of samples, at this CCP, met the standard (10 FC/g) compared to 86 % of cooked samples.

The lower performance of the approach to controlling FC contamination in Moni foods could be explained by the fact that, this food had low contamination levels before the intervention; indeed, more than 56 % of cooked Moni samples and 3% of reheated ones counted already less than 10FC/g before the intervention.

C) Intervention sustainability assessment

Health education programmes aim to induce behaviours promoting individual and community health. However this health promotion could be beneficial only when the new behaviours acquired become permanent. Following this concept, the intervention group mothers' capability to recall messages and to perform the corresponding corrective actions were assessed three months after the end of the intervention.

- **Messages recall assessment**

This step of the study aims to assess how long trainee mothers could remember messages learnt three months ago, considering that knowledge of the messages was the first step for their implementation.

Field workers were provided with a check list of all messages. A mother was asked to cite all messages she learnt and implemented during the intervention. The worker marked on the form messages recalled but neither asked or explained anything.

Table 3.16 below displays the number and the percentage of mothers who recalled accurately messages inducing actions to control FC contamination at CCPs.

Table 3.16: Message recall by 30 intervention group mothers, three months after intervention (using check list)

Message		N mothers recalling the message (%)	Observation
Use potable water for domestic needs		27 (90.0)	Water from tap or borehole
Wash dishes with water and soap		27 (90.0)	Local soap included
Wash two hands with running water and soap	After latrines	19 (63.3)	
	After cleaning child	11 (36.7)	
	After touching contaminated material	18 (60.0)	Including raw food
	Before preparation	26 (86.7)	
	Before child feeding	30 (100)	
Reheat leftovers to boiling point		30 (100)	Sometime after appearance of steam

Table 3.16 shows that the messages most recalled are hand washing before feeding a child and reheating, followed by using potable water for domestic needs and washing dishes with soap. Hand washing after cleaning a child registered the lowest score followed by washing hands after touching contaminated materials including raw foods.

• **Evaluation of the Intervention Group mothers’ ability to control foods’ FC contamination at CCPs**

Sampling at CCPs of both foods – Moni and Fish Soup – were undertaken again to assess the safety of these foods. Foods were sampled in the same conditions as during the intervention evaluation. Considering that mothers may not have the same behaviour in the presence and in the absence of field workers, the Intervention Group was divided into two sub groups of 15, for each food, during the assessment. 15 mothers’ foods were sampled during observational sessions and the 15 remaining foods were sampled without observation in order to find out the impact of field workers’ presence on mothers’ behaviour.

Table 3.17: Intervention Group FC count (FC/g) general trend in Moni at CCP three months after the intervention

Range(FC/g)	No. of samples (% of column total)			
	Intervention		Control	
	After cooking	After reheating	After cooking	After storage
ND	25 (83.3)	30 (00)	1(3.3)	0 (0.00)
10-100	5 (16.7)	0 (0.0)	4 (13.3)	1(3.3)
101-1000	0 (0.0)	0 (0.0)	25 (83.3)	27 (90.0)
1001-10 ⁴	0 (0.0)	0 (0.0)	0 (0.0)	2 (6.70)

N	30 (100)	30 (100)	30 (100)	30 (100)
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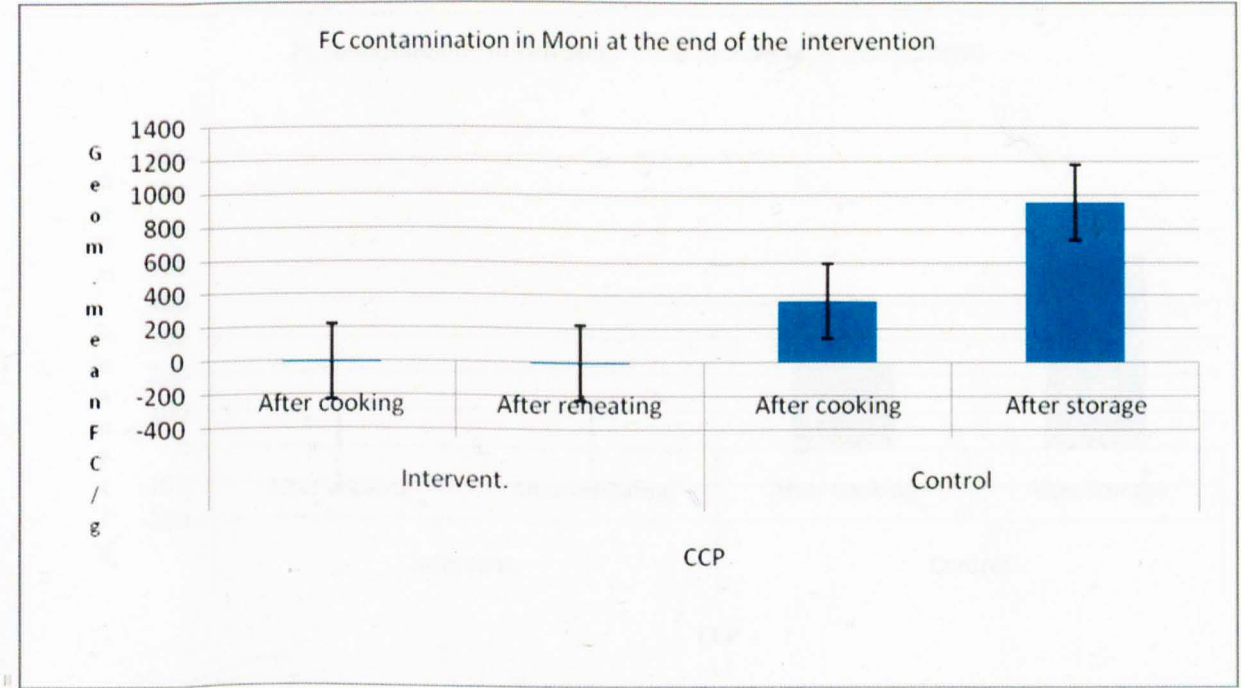


Figure 3.7: FC geometric means in Moni three months after the end of the intervention

Table 3.17 shows a FC contamination reduction three months after the intervention more important than that observed at the end of the intervention. Indeed, all reheated Moni samples met the standard set (10FC/g). But, after cooking FC contamination levels remained similar to that of the intervention. It is noticeable that the assessment occurred during the cold season when stored Moni seemed to be the most contaminated food; all stored food samples of the Control Group examined at this CCP counted more than 10 FC/g.

Table 3.18: Fish Soup Intervention Group FC count (FC/ g) trend at CCPs

Range(FC/g)	No. of samples (% of column total)			
	Intervention		Control	
	After cooking	After reheating	After cooking	After storage
ND	28 (93.3)	30 (100)	1 (3.3)	0 (0.0)
10-100	2 (6.7)	0 (0.0)	3 (10.0)	1 (3.3)
101-1000	0 (00)	0 (0.0)	26 (86.7)	28(93.4)

1001-10 ⁴	0 (00)	0 (0.0)	0(0.0)	1(3.3
N	30 (100)	30 (100)	30 (100)	30 (100)

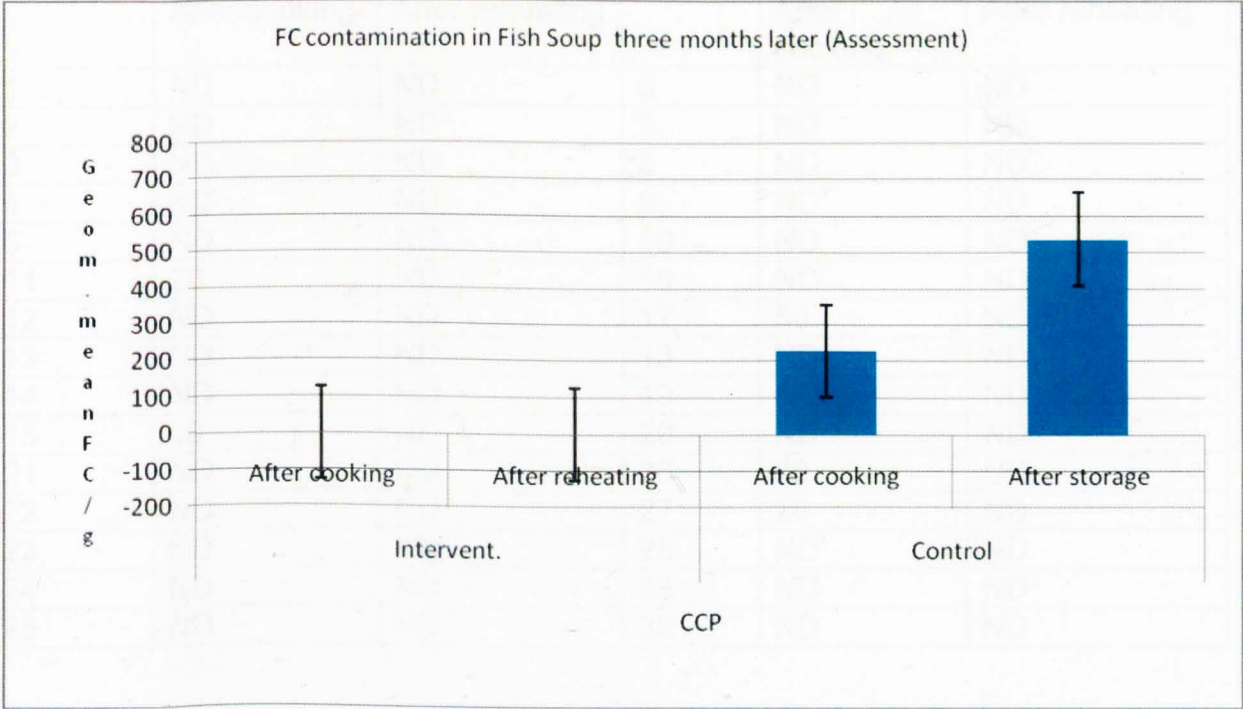


Figure 3.8: FC geometric mean in Fish soup three months after the end of the intervention

Table 3.18 records and Figure 3.8 indicate that the approach appeared to be more effective in FC contamination reduction in Fish Soup after cooking foods compared to those of the Moni. All reheated samples for both Moni and Fish Soup met the standard (10 FC/g). More than 93% of cooked Fish Soup samples met the standard whilst less than 84 % of cooked Moni samples met the same standard. Fish Soup assessment samples FC reduction appeared to be more important than those of the intervention. More than 93% of cooked samples and 100% of reheated samples of the former met the standard whilst less than 87% of cooked samples and less than 97% of reheated samples of the latter met the standard.

• **Observations’ impact on mothers behaviours**

This step aims to highlight FC count distribution between two sub groups resulting from the split of the Intervention Group. Households’ references are those used in the whole duration of the study therefore they are not in chronological order. One subgroup was observed during the assessment as in the intervention, and the second subgroup was not observed. Tables 3.19 and 3. 20 below display FC count for each sub group at CCPs.

Table 3.19: Moni sub groups' FC count (FC/ g)

Moni					
HHRef.	Non observed		HHRef.	Observed	
	After cooking	After reheating		After cooking	After reheating
1	ND	ND	6	ND	ND
2	ND	ND	7	ND	ND
3	ND	ND	8	ND	ND
4	ND	ND	9	ND	ND
5	ND	ND	10	ND	ND
11	20	ND	16	ND	ND
12	ND	ND	17	50	ND
13	ND	ND	18	ND	ND
14	ND	ND	19	ND	ND
15	10	ND	20	ND	ND
21	ND	ND	26	30	ND
22	ND	ND	27	20	ND
23	ND	ND	28	ND	ND
24	ND	ND	29	ND	ND
25	ND	ND	30	ND	ND

Table 3. 20: Fish Soup sub groups' FC count (FC/ g)

Fish soup					
HHRef.	Not observed		HHRef.	Observed	
	After cooking	After reheating		After cooking	After reheating
1	ND	ND	6	ND	ND
2	ND	ND	7	ND	ND
3	ND	ND	8	ND	ND

4	ND	ND	9	ND	ND
5	ND	ND	10	ND	ND
11	ND	ND	16	ND	ND
12	ND	ND	17	ND	ND
13	ND	ND	18	30	ND
14	ND	ND	19	10	ND
15	ND	ND	20	ND	ND
21	ND	ND	26	ND	ND
22	ND	ND	27	ND	ND
23	ND	ND	28	ND	ND
24	ND	ND	29	ND	ND
25	ND	ND	30	ND	ND

Data highlighted by the two previous Tables 3.19 and 3.20 indicate that on one hand, reheated food samples appeared to be less contaminated than cooked food samples, and on the other hand, non observed sub groups performed better than observed sub groups at all CCPs for both Moni and Fish Soup.

Table 3.21: Compararison of FC contamination levels of two weaning foods P values relative to the null hypothesis of no difference between observed and non observed mothers of the Intervention Group.

CCP	P
Moni	
After cooking	< 0.460
After reheating	Not applicable*
Fish Soup	
After cooking	< 0.171
After reheating	Not applicable*

Not applicable*: Geometric means are zero

There is no difference in controlling FC contamination at foods' CCP between observed and non observed mothers. It is noticeable that Fish Soup non observed sub group samples appeared to be less contaminated than those of the other sub groups.

- **Suspected risk factors for recontamination of food after cooking and reheating**

Despite the implementation of corrective actions during the intervention, some food samples of the intervention group remained contaminated. Then it became interesting to find out the impact of the potential environmental hazards identified during Flow Diagram construction (Tables 2.5 and 2.6) on cross contamination during the cooling step. The purpose was to check whether this recontamination was due to exposure to environmental hazards or to non-compliance with corrective actions. For

that purpose the RR related to the exposure to hazards was calculated for each food for contaminated samples.

Table 3.24 summarizes certain risk factors reported by field workers and supposed to be linked to this contamination; and Table 3.25 highlights the RR and their significance.

Table 3.22: CCP contaminated during the intervention and supposed risk factors related to this contamination

HHRef.	CCP		Risk factor
	Moni	Fish Soup	
11	After cooking		- Presence of flies
12		After cooking	- Presence of flies - pH>5
13	After cooking	After cooking	- Presence of flies - Poor environment ¹ - pH > 5 (Fish Soup)
14	After cooking After reheating		- Presence of flies - Poor environment ¹
16	After reheating		- Contaminated utensils - Presence of flies - pH>5
17	After reheating		- Presence of flies - Poor environment ¹ - pH> 5
20	After cooking		- Presence of flies - Poor environment ¹
23		After cooking	- Poor environment ¹ - pH> 5
24	After cooking		- Poor environment ¹
26	After cooking		- Poor environment ¹ - pH>5
27	After cooking		- pH> 5
30	After cooking		- Presence of flies - pH>5

¹ Presence in the household yard of one or both of the following hazards: waste water, solid waste or domestic animals (pet, goat, sheep, cow,...)

Table 3.23: Association between exposure to factor (potential source of contamination) and FC contamination of foods during the intervention.

Factor		RR	P
Flies		2.86	< 0.10
Poor environment		1.15	< 0.92
pH> 5	After cooling	1.92	< 0.46
	After reheating	1.63	< 0.84

Table 3.23 indicates that there is no association between factors of contamination and the contamination of food samples. These findings are very important as it

implies that the HACCP effectiveness is independent from the external factor, therefore the approach could be a very useful method to improving food safety in very hazardous environment like that prevalent in low income community settings.

3.. Study constraints and weaknesses

Three main constraints hampered the full development of this study. Firstly the very limited funds provided by the Malian Food Safety Agency, that as a developing country governmental organisation is facing a shortage of all kinds of resources. Secondly the limited capacity for bacteriological analysis of the National laboratory of Public Health which restricted the bacterial indicators to *faecal coliforms* alone, and the excessive cost of analysis that restrained the number of samples the present study could afford. Consequently the number of samples was limited to 432 during the experimental phase and to 720 during the pilot, to count only *FC*.

CHAPTER IV: Discussion

The present research findings are discussed under two headings, corresponding to the two phases of the study:

The two specific objectives of the **Experimental Phase** were: (i) to construct flow diagrams of the two foods mainly used as weaning foods in the study area and (ii) to test a small scale HACCP approach in which mothers' food preparation was supervised, to draw lessons on how to improve the microbiological safety of the two foods. The findings of the experimental phase of the study were reviewed in accordance with the HACCP general approach and compared to similar studies, to ascertain whether the specific objectives mentioned above were met.

The principal objective of the **Pilot Study** was to take the experiment nearer to reality by using the experiment outcomes (delivery of messages) to improve food safety. Specifically, it was to eliminate or reduce to an acceptable level, at CCPs, FC contamination in weaning foods through the delivery of messages at household level, and to develop recommendations on how to apply the HACCP approach to domestic food preparation in developing countries. This phase was reviewed focusing on its effectiveness and its sustainability, comparing FC contamination outcomes in the Intervention Group before and after the intervention, whilst the Control Group ones helped to follow foods bacteriological contamination over the seasons. The results showed the relevance of the HACCP approach to domestic food preparation and handling in a very low income community in developing country, and confirmed its applicability which had been predicted by previous studies (Bryan, 1992; Ehiri et al, 2001).

The HACCP approach is widely used in food manufacture, international import and export trade channels, in supermarket supply systems and management as well, in developed countries because it is considered as one of the most effective tools for protecting food safety. The system was particularly adapted to these above mentioned environments because the teams involved are very highly skilled, well trained and well led, working in a very limited and controlled space so that it is very easy to fulfil HACCP preliminary hygiene requirements. On the contrary, at household level, especially in developing countries, the household environment in general, and kitchen environment in particular, is very poor because of the lack of cleaning, the presence of domestic animals and the ignorance of family members about hazards related to food borne disease transmission. Then implementation of the HACCP approach at home needs some flexibility that is fortunately admitted in its principles (Bryan, 1992; Ehiri et al, 1995). That flexibility was fully used in the experimental phase of the present research to overcome some differences between home settings and very well controlled industrial environments.

4.1 Experimental phase

Most of the experimental study sample of 15 volunteer mothers, with children aged between 6 and 16 years old, was young. They were all aged less than 35 years old. More than half was illiterate (about 50% were non-schooled); more than 53 % were

house wives without independent income. The figures for the fathers were similar, except age; they were on average 10 years older; about 49 % were non- schooled and more than 53 % were artisans or drivers (Table 2.1). In consequence, according to the criteria of the Malian National Bureau of Statistics and the Census, these households pertained to the low income class.

Regarding mothers' traditional behaviour, 12 out 15 declared washing their hands before touching foods; 14 declared doing the same after using a latrine; 14 checked the temperature of cooked foods with their hands before feeding their child ; 14 fed their child from a plastic cup and 2 used their hands (Table 2.4).

As advised by the HACCP approach guidelines, the experiment team were diverse with a variety of specialists in fields related to food safety. Indeed all members were employees of the national Food Safety Agency, the governmental body in charge of coordination of food safety activities at national level. Field workers were all university graduates and mothers involved were very committed volunteers. Several meetings with the volunteers and other actors ensured the wide participation of all interested parties in the planning of the experiment. This is consistent with the principles of the HACCP approach (Bryan et al, 1992; CAC, 1993; NSF, 2006).

Ideally all foods prepared at home must undergo the HACCP approach in order to protect consumers' health, however home foods are very diverse due to many factors (for example, members choice, cultural habits and family income) so that it is not easy to cover all of them. Fortunately, in all communities or countries, mainly in low income groups there is a main recipe, basis of the most consumed meal (Table 2.3 and 2.4). In addition, the type of foods, economic resources and cultural influences often result in considerable uniformity within groups or sub groups of the society (Bryan, 1992). Some meals like rice based meals are even adopted in different continents. It is also recognised that infant and children's foods are mainly prepared and stored within homes, (Esrey and Feachem, 1989). Thus in this experiment, we selected the two meals mostly used as weaning foods, during unstructured focus groups gathering volunteer mothers (Table 1.3).

Describing the processes of food preparation and handling is a crucial step, as it is the basis of the flow diagram so that any failure at this step could result in an insufficient flow diagram that in turn could negatively impact the following steps. Many tools are needed to collect comprehensive and accurate data (Bryan, 1992). Different focus groups, environmental walks (to visit household premises, local markets, and private taps and boreholes) and observations of food preparation and handling were conducted to list information related to raw products and processes of preparation and handling of the two foods (tables 2.3-2.6). The scrutiny of this step helped to find out that some operations, though claimed, didn't in fact occur, confirming that an individual's knowledge and their actual practice are not always consistent (Lanata et al, 2003).

Intended use of weaning foods is quite simple to find out because the target group, children aged less than 16 months, is well established.

Figures 2.4 and 2.5 represent respectively the Moni and the Fish Soup flow diagrams. Each food had its specific flow diagram depending on its raw products, the processes it undergoes, and the handling patterns. But sources of potential

contamination identified for both Moni and Fish Soup were comparable because of their dependence on the same environmental hazards (Figures 2.4 and 2.5) and were consistent with those found by many similar studies conducted in developing countries (Bryan, 1992; Sheth et al, 2000; Ehiri et al, 2001). Most food preparation and handling operations were exposed at the same time to all of the potential hazards listed (Tables 2.5 and 2.6), showing the very high exposure of food to contamination in low income settings (Madhu et al, 1996).

The Moni flow diagram is the more complex due to the diverse raw material used and the high number of operations involved resulting in occasional practice of certain operations (flour cooking) or complete omission of some practices known but not actually implemented. The latter was due probably to mothers' lack of time or resources, or simply because the practice (reheating) was learnt but never practised. Ghuliani (1995) found in Bangladesh that occasional reheating (lukewarm during 2-3 min or boiling for 5-7 min) practised by mothers was not effective in pathogen elimination. The occasional practice of flour cooking was highlighted, but the operation of reheating was omitted in the diagram, as the latter must reflect actual practice (Bryan, 1992; Mitchell et al, 1992; CAC, 1993).

The hazards analysis confirmed the presence of bacteriological hazards at all suspected preparation and handling operations except for two raw materials (maize and home piped water), adding ingredient (Monkey bread) and cooking. Monkey non contamination could probably due to the fact that this ingredient is not only used to sweeten foods in replacement of sugar (more expensive), but also as a preservative for flour stored at least during one week. Henry (1990) found that adding salt to left overs caused considerable multiplication of *vibrio cholerae* within 24 hours.

Contamination of raw materials ranged between respectively 20-310 FC/ g for water (water provided by vendors was one of the most contaminated), 20- 1000 FC/g for cereals (millet, the most common and the cheapest, thus the most affordable was the most contaminated), 50- 130 FC/g for the flour mixed with water (the first step of making Moni), 110-840 FC/g for the fresh fish and 20- 1000 FC/g for vegetables; the two latter were used in Fish Soup preparation (Table 2.9). These levels of contamination were consistent with those listed in Table 1.1 summarizing the findings of similar studies.

It is noticeable that cooking was very effective in FC elimination; indeed no cooked food sample contained this germ, due probably to the high temperature of cooking, 91.1°C during 22 min for the Moni food and 87.8°C during 40 min for the Fish Soup, far above the requirement of 60°C during 12 min (Bryan, 1992). These results were consistent with those mentioned in cooking temperature curves (> 70°C) of various foods in a village and an urban setting in developing countries, reported in Bryan's (1992) review. But recontamination occurred, mainly in Fish Soup during storage time (Table 2.10 and 2.11); this is consistent with what Henry (1990) found in rural Bangladesh. He observed that stored foods were more contaminated than those eaten earlier.

Some samples of all foods were contaminated after storage at room temperature (33.2-36.6 °C) for more than 6 hours. Two Moni samples were contaminated (120 and 290 FC/ g) and 9 out of 15 Fish Soup samples contained between 30 and 550

FC/g. Moreover, Fish Soup was as contaminated as the fresh fish ($P < 0.0294$). Cooked food leftovers stored at room temperature more than 2 hours were found highly FC contaminated in all related studies summarized in Table 1.1.

Following the decision tree scheme (Figure 1.2), 4 and 5 CCPs were found respectively for the Fish Soup and the Moni. But the FC elimination advantages of the first Moni CCP (flour cooking) were completely lost during weeklong storage at room temperature in contaminated containers (empty manufactured milk cans). This operation is then a waste of the very scarce time and resources of poor communities.

In addition, the attention needed to control hazard at CCPs, is so time and resources consuming that the fewer these CCP are the easier could be their control. Moreover, the Moni cooking was very effective as indicated by the very high temperatures and durations reached (91.1°C - 22 min) during the whole experimental period as found by Bryan's (1992) review. As a result, the flour cooking CCP was abandoned.

FC contamination occurred in both Moni and Fish Soup, during their storage, producing FC counts up to 550 FC/g, making storage the most hazardous step. Henry (1990) found in rural Bangladesh that there was no difference in contamination between covered and uncovered stored foods. Thus, to make leftover safer for consumption, the only way is to introduce reheating as no hygiene or sanitation measure would have an effect on the contamination at this step. This corresponds to conditions of the HACCP decision tree for which no preventive measures exist (question 1 answer is no) at a step at which contamination control is necessary for safety; it was recommended to modify the step in order to introduce a CCP.

Accordingly, a modification at this step is needed; as a result, reheating was introduced as a CCP. **In total, 4 CCPs for each food were definitely adopted: cooking, reheating and serving after cooking and after reheating** (Figures 2.9 and 2.10). Bryan (1992) mentioned in his review 3 CCPs for canned milk: boiling water, milk-can opening and storage. Sheth (2000) found 2 CCP for Chappati foods: roasting and holding at ambient temperature. Ehiri (2001) found 3 CCPs for Jollof rice, Moi-moi and Agidi, including cooking and reheating, and 2 CCPs for Ground crayfish including handling. Table 1.2 summarises CCPs found in selected studies' findings. The common results of all these findings are CCPs at the cooking and reheating control points. The other CCPs: holding leftovers at room or ambient temperature or storage, recommended, instead as control measure by Ehiri (2001), are subject to caution because no effective measure is available in replacement of reheating. Other measures proposed, such as covering foods, were found not to be effective (Henry et al, 1990).

Obviously the temperature and time chosen as critical limits for cooking and reheating CCPs must be higher than (60°C - 12 min) (Bryan et al, 1992), thus 70°C and 10 min were selected. It is important to underline that the critical limits selected were less than cooking temperatures recorded during the present experiment sample mothers' traditional cooking observations. Except for the review conducted by Bryan (1992) no study listed in Table 1.1 proposed critical limits, as none of them continued the HACCP approach beyond setting CCPs. To control FC contamination at the two remaining CCPs, serving the child after cooking and after reheating, two behavioural control measures were selected: washing hands with potable water (water from a

home tap or borehole) and soap at critical periods (after using latrines and cleaning a child's bottom, and before handling foods), and cleaning utensils with potable water and soap (including local soap). Ehiri (2001) recommended the same measures for the 5 foods listed in Table 1.1. But Bryan (1992) in his review mentioned three control measures for this kind of CCPs: prompt service/eating, cooling rapidly in a shallow pan or holding hot ($>55^{\circ}\text{C}$). Obviously the two latter are not applicable in this studied community, where heating or cooling equipments are not available (table 2.2).

To monitor CCPs, in accordance with the critical limits selected, temperature and time measurement immediately after cooking or reheating were chosen for cooking and reheating CCPs and observation of practices (handwashing and cleaning utensils) were selected for child feeding/service with cooled foods. Bryan (1992) in his review mentioned the same measures for similar foods. Instead, Ehiri (2001) recommended different monitoring system like cooking and reheating thoroughly, eating promptly after cooking or reheating, and protection of the food against certain sources of contamination (dust, flies etc..). These latter were considered as control measures in Bryan's (1992) review and in the present study. Ehiri (2001) did not recommend critical limits in his study, as the latter did not continue beyond identification of CCPs.

As highlighted in Table 2.12 and Table 2.13, the following temperature ranges were recorded: 94.1°C - 99°C and 86.7°C - 98.7°C respectively for cooking and reheating of Moni, 89.5°C - 99.3°C and 77.8°C - 99.7°C respectively for cooking and reheating Fish Soup. In the same order the following ranges of time duration were recorded: 10 min - 20min and 04 min - 11min respectively for cooking and reheating Moni, 9 min - 65 min and 4min- 25 min respectively for cooking and reheating Fish Soup. All temperatures recorded were far above the critical limits. Deviation from critical limits was observed in terms of duration at both cooking and reheating CCPs for both Moni and Fish Soup. Indeed, all durations less than 12 min were recorded as deviations.

The highest temperature (99.3°C) was observed, and the longest duration (65 min) was also recorded, at the same Fish Soup cooking CCP. The lowest temperature (77.8°C) and shortest duration (4 min) recorded were at reheating CCPs for Moni and Fish Soup. In general cooking parameters were higher than the reheating ones for both Moni and Fish Soup. Fish Soup took the longest cooking duration. These differences could be attributable to the fact that certain raw products need more time and temperature to be well cooked. Fresh fish, an ingredient for Fish Soup, requires a long cooking time. Reheating of leftovers, targeted only at FC elimination in foods, requires less time. However, it was effective; no FC were found in cooked and reheated food samples as shown in Table 2.14 and 2.15. This suggests that time deviations were compensated by the very high temperature recorded (Bryan, 1992). Monitoring practices produced reports of all behavioural deviations. Corrective actions were recommended to mothers immediately after the occurrence of a deviation, and hand washing kits (equipment and soap) (Figure 2.13) were distributed to mothers.

One corrective action was common to cooking and reheating, heating foods at least to the critical temperature and time (70°C – 10 min); the other corrective actions were, washing hands with potable water and soap at critical periods (after using latrines or cleaning a child's bottom, or before handling foods), washing utensils with

potable water and soap, and covering foods. Bryan (1992) mentioned in his review similar corrective actions for similar foods. They are also included in the control actions recommended by Ehiri (2001) for 5 foods listed in Table 1.1. Tables 2.14 and 2.15 display the monitoring results based on FC count of foods sampled at CCPs. No FC were found in immediately cooked or reheated food samples. Only two Fish Soup samples at serving CCP were found positive with 72 and 1000 FC/g.

All in all, cooking was found very effective in FC elimination at corresponding CCPs in both Moni and Fish Soup ($P < 0.0001$). Reheating learnt and regularly adopted by mothers only during the experiment became as effective as cooking ($P < 0.0001$); this is confirmed by the absence of a significant difference between cooking and reheating temperatures of both Moni and Fish Soup, $P < 0.026$ and $P < 0.071$, respectively.

In conclusion, the two specific objectives of the experimental phase were fully achieved and the study was taken nearer to reality throughout the second phase.

4.2 Pilot Study

The 2 samples of the Pilot Study – 60 mothers with children aged between 6 and 9 years old – were similar to that of the Experimental Phase. They were young (less than 35 years old), more than 56 % were illiterate, about 50% were housewives without independent income. Males presented similar figures; about 39 % were illiterate; most of them were workmen (Tables 3.1 and 3.2). Therefore, these households pertained also to the low income class and represent the same group as the subjects of the experimental phase.

Baseline physical parameters of foods, Moni and Fish Soup, showed that cooking was very effective for FC elimination with cooking temperatures above 97°C and duration time at least 4 min. The temperatures of foods served to children after cooking and after storage ranged between 28.4°C and 48.3°C . No case of reheating was observed. Foods were stored for at least 2 hours and sometime more than 6 hours. All foods had a pH less than 7 (Table 3.3). No significant difference in baseline physical parameters was found between the Intervention and the Control Groups ($P < 0.23$ to 0.76), (Table 3.4).

As traditional cooking was confirmed to be very effective in FC elimination, no sample was taken immediately after cooking or after reheating, for FC count. Thus FC were counted only at two CCPs: service after cooking and service after reheating, named respectively, After cooking and After reheating.

Baseline FC contamination levels ranged between 10 and 10000 FC/g for two foods into two groups (Tables 3.5 and 3.6). These FC levels are similar to most of the findings of previous studies listed in table 1.1.

No significant difference in FC contamination levels was found between dry and rainy seasons at CCPs after Moni storage, after Fish Soup cooking and storage, or between dry and cold seasons at Moni cooking CCPs and after Fish Soup cooking

and storage CCP, as between rainy and cold seasons at after Moni cooking and storage and after Fish soup cooking ($P < 0.290$ to 0.720). A significant difference in FC contamination levels was found between dry and rainy seasons at CCPs after Moni cooking ($P < 0.0004$), between dry and cold seasons at the same CCPs ($P < 0.0002$), and between rainy and cold seasons at CCP after Fish Soup storage ($P < 0.0098$). Where a difference was found, the level of contamination was higher in December followed by the rainy season; the dry season recorded the lowest levels of contamination at all CCPs (Table 3.7). Henry (1990) and Imong (1995) found, in rural Bangladesh and rural northern Thailand respectively, that contamination was higher in the rainy season compared to the dry season for most of the foods they studied.

There was a significant difference in contamination levels between the two CCPs: after cooking and after storage for the two foods. For both Moni and Fish Soup, the CCP after storage was more contaminated than that of after cooking, ($P < 0.0001$) (Table 3.8). This is consistent with the relation between microbial growth and the couple temperature-time; indeed, at suitable temperatures, the longer the storage time the higher is the microbial contamination.

It could be concluded that, in spite of the opportunity for the Control Group mothers to learnt from their counterparts in the Intervention Group, their food contamination patterns remained similar over the seasons of the year.

An evaluation of messages understood after three weeks' training showed surprisingly that reheating, though it was not mothers' traditional behaviour, was the most understood and implemented (100%), and followed by potable water usage for domestic needs (90%) and washing of dishes (26.67%). Hand washing with potable water and soap at critical periods scored less (27%) than the other behaviours (Table 3.9) though 12 out of 15 experiment group mothers living in the same area said they always washed their hands before touching foods and 14 out of 15 said they did so after using a latrine (Table 2.4). In a trial study, five messages (hand washing before and after defined events, boiling water for reconstituting of powdered milk, feeding gruel by spoon rather than bottle-feeding, not storing gruels and milks, and all four together) were delivered to groups of 15 mothers pertaining to families living in poverty and resulted in the adoption and the advocacy of corresponding behaviours by 53% to 80 % of mothers, and their being practised every time during one-month period (Monte et al, 1997).

A comparison between FC contamination levels recorded during the intervention evaluation of Intervention Groups (Table 3.11 and 3.13) and their baseline data (Tables 3.10 and 3.12) for two foods at the end of the intervention showed a very significant difference ($p < 0.0001$). This difference is less important at Moni cooking CCPs ($P < 0.04$) (Table 3.15). Indeed at these latter CCPs baseline contamination levels were very low even before the intervention; more than 56 % of them counted less 10 FC/g (Table 3.10).

The intervention was very effective in FC contamination reduction; therefore the HACCP approach was very effective.

However, the fact that all intervention mothers were volunteers could imply that they were already dedicated to behavioural changes and committed to learn more

because they knew the advantages and/or they were able to afford corrective actions (e.g reheating).

4.3 Relative reductions in bacterial numbers

One of the most important advantages of the HACCP approach is its ability to help meeting the requirements of standards. The intervention performance (the ability of the intervention to ensure foods safety) was assessed with regard to intervention and control group FC counts meeting the standard fixed at the onset on this phase (10 FC/g),. Very high performance was recorded at all CCP.

The Relative Risks (RR) calculated were higher than 1, indicating that the risk of not meeting the standard was higher in the Control Group compared to the Intervention one. RR is 5.8 and 4.8 at Moni cooking and reheating CCPs respectively, and 6.8 and 15 at Fish Soup CCPs (Table 3.16). The highest performance was recorded at Fish soup CCPs that were the most contaminated before the intervention in all groups through the seasons (Tables 3.5, 3.6 and 3.12) and during the experimental phase (Table 2.11). For two foods the intervention was more performing at reheating CCPs than cooking even though cooking and reheating were found equal effective. The lower performance at Moni CCPs compared to the Fish soup ones could be attributable to a lower level of contamination before the intervention (Table 3.10).

The intervention resulted in a very high performance in meeting the quality standard of less than 10FC/g; in consequence the HACCP approach improved the microbiological safety of both foods very significantly.

4.4 Persistence of the behaviour change

The adoption of new behaviours is important in health education programmes, but the most important is their sustainability; thus the intervention's impact was assessed three months after the end of the intervention. Message recall showed again that reheating was the most remembered message (100%) together with hand washing before feeding the child (100%) and followed by potable water usage and dish washing (90%); washing hand at certain events recorded the lowest score: after using latrines (63.33%), before food preparation (60%) and after cleaning a child's bottom (36.66) (Table 3.16). These results were consistent with those of the intervention training assessment (Table 3.9) where reheating was the most understood and practised message followed by potable water use and dish washing; hand washing was the least understood and implemented message.

A comparison between FC contamination levels at CCPs recorded for the intervention baseline and those recorded three months after the intervention resulted in a greater reduction of these levels. Then the assessment period FC reduction became more effective than that of the intervention, ($P < 0.0001$) at all CCPs even for Moni after cooking ($P < 0.004$).

Behaviours acquired lasted for at least three months after the intervention.

4.5 Effect of observations (“Westinghouse effect”); risk factors

As mothers were observed during the whole study till the end of the intervention, the impact of this observation was also assessed by comparing FC levels in their food samples with those in a sub-group of 15 intervention mothers whose food preparation had not been subject to observation. No difference was found in microbial food quality between the two sub groups, ($P < 0.450$) at Moni cooking CCP, and ($P < 0.171$) at Fish Soup cooking CCP (Table 3.21).

As a result it could be inferred that observation by field workers had no impact on mothers' behaviour.

The relationship between certain risk factors identified (Table 3.2) during households' socioeconomic data collection and the persistence of the contamination of some food samples despite the intervention, was checked. No difference was found between food samples exposed and not exposed to these factors (Table 3.22). These findings could have important implications for the future of the HACCP approach in improving home food safety. Indeed an independence of the HACCP efficacy from environmental factors implies that the approach could improve foods safety even in a very hazardous environment like that of the low income communities in developing countries.

4.6 Appropriateness of the HACCP Approach in Low Income Community Settings

The HACCP approach, to be relevant for home food safety in low income community, must be flexible, scalable, affordable, feasible and acceptable.

Flexibility

Given that foods were consumed locally, CCPs could be reduced to the most relevant of them, instead of considering all in accordance with the HACCP decision scheme. Following this latter, at each control point where a preventive control measure exists and if this step is specifically designed to eliminate or reduce the likely occurrence of hazard to an acceptable level, then this control point must be considered as a CCP.

As an example, in Moni preparation, flour cooking is very effective, so that in accordance with the HACCP decision rules, the control point at this step is automatically a CCP. However, all advantages gained for FC elimination were lost during the drying and storage steps; moreover, Moni cooking, a subsequent step, was very effective in elimination of bacteria. Thus, the flour step CCP was abandoned in this present study and no negative impact resulted for Moni safety. This flexibility of the HACCP approach is already recognised (Bryan, 1992; CAC, 1993; Ehiri et al, 1995, 1996; WHO, 1999). The reduction of CCPs in home foods to the minimum possible is a key factor in its success. The fewer CCPs, the easier is their control, which demands time and resources.

Scalability

Contrary to what one might imagine, there is not considerable variation in food preparation practices in individual homes, because food types, energy resources, cooking facilities, incomes and traditional and cultural influences, often result in considerable uniformity within groups or sub groups of a society (Bryan, 1992). This is more likely for a well-targeted group like children in very poor rural or peri-urban communities in developing countries. Indeed it is recognised that infants' and children's foods are mainly prepared and stored within homes, (Esrey and Feachem, 1989). In addition, certain raw products, like rice, wheat or maize are widely used in all continents. Their preparation and handling follows the same procedures, at least in each continent. Therefore, the HACCP approach to hazard analysis could result in common information associated with preparation, storage and handling of foods, to assess risk and to identify critical control points. Ehiri (2001) found 3 CCPs (cooking, storage, and reheating) common to the 5 foods he studied, including rice, as in the present study. The same three CCPs were identified for rice in Bryan's (1992) review; Sheth (2000) found two of them (cooking and reheating) as CCPs for Chappati, an Indian local food for which rice is a component.

In very deprived rural and urban communities like those of Mali, the site of the present study, scaling up the HACCP approach at household level could be hampered by the very limited resources (money, potable water sources and even time); however this research has shown that very high commitment of the stakeholders could help to overcome these obstacles.

Affordability

Hazard analysis, one of the most important steps of the HACCP approach is expensive and time consuming, as analysis of food samples is involved, but it is carried out only once, and results are usable not only in the community for which it is intended but also elsewhere for the same food or other food undergoing the same processes. In addition, the identification of associated hazards at any stage of food preparation and handling, the assessment of relative risk and the determination of CCPs, where control is effective, helps to focus on hazards control at these steps (CCPs), instead of traditional inspection procedures demanding the surveillance of all steps of the food preparation and handling. Moreover, the monitoring commonly practised by food inspectors in public places is often impracticable in home.

However, for very poor peri-urban African residents, the cost of fuel wood or charcoal could hinder the sustainability of reheating as a corrective measure to improve the bacteriological quality of stored foods.

To maximize the HACCP application and its impact in health promotion, hazard analysis should be conducted in regions with the highest incidence of diarrhoea, in families with children with history of diarrhoea. Moreover, in regions with high infant mortality, households with children of weaning age should be targeted.

Feasibility and acceptability

Most health education programmes limit themselves to raise public awareness and messages delivering, to motivate behavioural change which could result in actions, for instance to control foods contamination and improve their safety. Messages

advocating behavioural changes which require expenditure of resources (money, time or energy), like reheating, boiling water or getting soap, compete with vital activities, and so are generally ignored (Henry et al., 1990). If, only food sufficient for one meal is to be prepared at a time, this requires that food be prepared and cooked more often; this may be more expensive, even impractical, (Esrey and Feachem, 1989); the present study found that reheating took less time than cooking to reach the same temperature. Esrey and Feachem (1989), in their review of interventions for promotion of food hygiene, argued that a well designed program of messages delivering about food hygiene to mothers could lead to changes in food hygiene in the home. Monte (1997) designed and implemented five educational messages to improve weaning food hygiene practices of mothers living in poverty in an urban slum in Fortaleza, Brasil, and achieved the targeted behavioural changes and their sustainability during a one month period, despite mothers' limited material resources. The present study found also that reheating, which was not a traditional habit of our sample of mothers, became the most remembered message and the most implemented practice, and reheating stored foods become the safer step. These two findings (Monte et al, 1997 and this present research) are starting points to confirm Esrey and Feachem's hypothesis.

Chapter V: Conclusions and recommendations

5.1 Conclusion

The present research showed not only that the HACCP approach is effective in improving home food safety but also that it is relevant for food hygiene and safety promotion in a low income community. Indeed the experiment resulted in a dramatic reduction of FC at all CCPs (cooking, reheating and cooling before child feeding); and these findings were confirmed by the intervention which demonstrated that the HACCP approach significantly improved the bacteriological quality of foods prepared by the Intervention Group mothers in a very harmful environment during all seasons of the year.

Not only did the HACCP approach result in an improvement of the bacteriological quality of foods, but also behaviours acquired by the Intervention Group mothers lasted and even improved three months after the intervention ended. On the other hand, the effectiveness of HACCP seems to be independent from environmental hazards, which could be most interesting for the future of the approach in the poor settings of developing countries.

5.2 Recommendations, including questions for further research

Esrey and Feachem (1989), in their review of interventions for promotion of food hygiene, argued that for the promotion of food hygiene to be proved to be an effective diarrhoea control measure, either the first two or the third of the following hypotheses must be confirmed as true:

- 1) poor food handling, preparation, and storage practices contribute to the transmission of diarrhoea-causing pathogens and thereby increase diarrhoeal morbidity or mortality rates among young children; and
- 2) food handling, preparation, and storage practices can be improved by appropriate educational and legislative measures; or
- 3) appropriate educational and legislative measures to improve food handling, preparation, and storage practices can reduce diarrhoeal morbidity or mortality rates among young children.

Whilst studies of the first hypothesis are still few and contradictory, some findings, Monte (1997), and the results of the present study give hope that the second and the third hypotheses are true. However to prove it, important research questions need to be answered:

Firstly, could food safety improvement, achieved through the HACCP approach, result in diarrhoea morbidity and mortality reduction among young children?
Secondly, is the approach scalable and cost effective?

Research resulting in a positive answer to these two questions could be an extraordinary achievement in infant diarrhoea prevention, as it has been recognised that food plays a much more significant role in the transmission of enteric diseases than previously thought (Kaferstein, WHO Bull. 2003). Thus, food safety could take its rightful place in the strategy to prevent infant diarrhoea, beside (i) the promotion of breastfeeding, (ii) vaccination against certain childhood diseases and (iii) improved water supply and sanitation.

5.3 Suggestions for new research implementation

The next step of this research could aim to find out the scalability of the HACCP approach at household level in a developing country and its impact on infant diarrhoea prevention or reduction. For this purpose, it could be useful to take advantage of the presence of local radio stations broadcasting messages in local languages, in many developing countries (Malian villages are covered by more than 600 local radio stations). In order to reduce the bias related to the influence of television, it is possible to select villages with local radio which are not covered by the TV. In selected villages mothers of children with a history of diarrhoea will be considered. One group of villages will receive HACCP educational messages through the local radio, and the second group not receiving messages. The two groups of villages must be far from each other in order to avoid the impact of messages on the control group villages.

REFERENCES

- Adak G.K, Meakins S.M, Yip H, Lopman B.A., O'Brien S.; Diseases Risks from Foods, England and Wales, 1996-2000; *Emerging Infectious Diseases*;2005, Vol 11, No 3:365-372.
- Adams M.R. and Moss M.O.; Food Microbiology, second edition, *Royal Society of Chemistry*, London, 2000.
- Afifi ZE,Nasser SS., Shalaby S., Atlam SA; Contamination of weaning foods: organisms, channels and sequelae; *Journal of Tropical Paediatric.*; 1998 dec,44(6):335-7.
- Al-Ali F.M., Hossain M.M.and Pugh R.N.; The association between feeding modes and diarrhoea among urban children in a newly developed country; *Public Health*,1997 ,111(4):239-43.
- Baker J., Naeeni M., Bloomfield S F.; The effects of cleaning and disinfection in reducing Salmonella contamination in a laboratory model kitchen; *Journal of Applied Microbiology*; 2003, 95:1351-1360.
- Banwart G. J.; Basic Food Microbiology; *AN Avi Books*; New York; 1995.
- Bartram J.,Corrales L., Davison A., Deere D., Drury D., Gordon B., Howard G., Rinehold A., Stevens M.; Water Safety Plan: Step-by step risk management for drinking water suppliers; *WHO/IWA*, Geneva, 2009.
- Bjorn L; Household Water Supply, Sanitation, Hygiene and Diarrhoeal Disease in Children: An Analysis of the Senegal Demographic and Health Survey 2005; *A Background Report for the Senegal CEA World Bank*; Dakar, 2007.
- Bloomfield S B.; Home Hygiene: a risk approach; *International Journal of Hygiene and Environmental Health*, 2002, 205, 1-8 .
- Bloomfield S B, Nath K.J.; Home Hygiene in Developing Countries Prevention of infection in the home and the peri-domestic setting: A training resource on hygiene for teachers, community nurses, community workers and health professionals in developing countries; Water Sanitation and Hygiene, *International Scientific Forum on Home Hygiene*,2006 , 157p.
- Boschi-Pinto C., Velebit L., Shibuya K.; Estimating Child Mortality due to diarrhoea in developing countries; *Bull. WHO*; Geneva; 2008, 86(9):710-7.
- Bryan F.L.; Hazard Analysis Critical Control Point Evaluations: A guide to identify hazards and assessing risks associated with food preparation and storage, WHO, Geneva, 1992.
- CAC; Procedural Manual; Rome: 17th edition, Codex Alimentarius Commission 2008.

CAC; Recommended International Code of Practice- General Principles of Food Hygiene, Rome: Codex Alimentarius Commission, rev4-2003.

Clayton D.A, Griffith J. C; An investigation of the factors underlying consumers' implementation of specific food practices; *British Food Journal*, 2003, 105 (7):434-453.

Clayton D.A, Griffith J. C; Observation of food safety practices in catering using national analysis; *British Food Journal*, 2004, 6 (3):211-227.

Cellule Sectorielle de Lutte contre le Sida; Rapport 2009; Ministere de la Sante.

Cogan T.A., Slader J., Bloomfield S.F. and Humphrey T.J.; The effectiveness of hygiene procedures for prevention of cross-contamination from chicken carcasses in the domestic kitchen; *Letters in Applied Microbiology*; 1999, 29:354-358.

Cogan T.A., Slader J., Bloomfield S.F. and Humphrey T.J.; Achieving hygiene in domestic kitchen: the effectiveness of commonly used cleaning procedures, *Journal of Applied Microbiology*, 2002, 92,885-892.

CPS/MS (Cellule Planification Statistique/Ministere de la sante/MALI) ; Enquete Demographique et de Sante ; Rapport Preliminaire ; Avril 2007.

Curtis V. and Cairncross S., Effect of washing hands with soap on diarrhoea risk in community: a systematic review, *Lancet Infectious diseases*, 2003:3:275-81.

Curtis V., Cairncross S. and Yoni R.; Domestic hygiene and diarrhoea – pinpointing the problem, *Trop Med Int Health* 2000:5:22-32.

Curtis V. and Kanki B.; Planning a hygiene promotion programme; Risk practices, target practices ; Motivating behaviour change ; Communicating hygiene; New York; 1998.

Cutting W. and Elliott K. ; Making weaning safer; *Dialogue on Diarrhoea*, 1994 ;(56):1.

Ehiri J .E, and Morris G.P; Food safety control: Overcoming barriers to wider use of hazard analysis; *World Health Forum*; 1996, 17; 301-303.

Ehiri J E., and Morris G.P; HACCP implementation in businesses: the need for a flexible approach; *Journal of the Royal Society of Health*; 1995:995-253.

Ehiri J E. and Prowse J.M.; Children health promotion in developing countries: the case for integration of environmental and social interventions? *Health Policy and Planning*; 1999, 14 (1): 1-10.

Ehiri. J E, Azubuike.M C, Ubbaonu.N C et al; Critical Control Point of complementary Food Preparation and handling in eastern, Nigeria; WHO Bulletin; 2001; 79;423-433.

Ersey S.A. and Feachem R.G.; Intervention for the control of diarrhoeal diseases among young children: Promotion of food hygiene, WHO, Geneva, 1989.

FAO/WHO: Guidance to governments on the application of HACCP in small and/or less-developed food businesses; *Food and Nutrition paper* 86.

Ghuliani A. and Kaul M. ; Contamination of weaning foods and transmission of *E. coli* in causation of infantile diarrhoea in low income group in Chandigarh; *Indian Journal of Paediatrics*, 1995, 1- 32 .

Gilling S.J., Taylor E., Kane K., Taylor J.Z.; Successful hazard analysis and critical control point implementation in the United Kingdom: understanding the barriers through the use of behavioural adherence model; *J. Food Prot.*; 2001, 64(5):710-715.

Griffith J. C; Worsfold D.; Application of HACCP to food preparation practices in domestic kitchens, *Food Control*, 1994, vol 5, N 3.

Haggerty P A., Kalengaie M., .Kirkwood B.R., Ashworth A and Manwela M.; Methodological Approach in a Baseline Study of Diarrhoeal Morbidity in Weaning – Age Children in Rural Zaire; *Journal of Tropical Medicine and Hygiene*;1994:1040-1049.

Haggerty P A., Kalengaie M., .Kirkwood B.R., Ashworth A, and Manun'ebo M. N.; Community –based Hygiene Education to Reduce Diarrhoeal Disease in Rural Zaire: Impact of the intervention on the Diarrhoeal Morbidity, *International Journal of Epidemiology*, 1994; 23, 1050-1059.

Haggerty P A., Kalengaie M., .Kirkwood B.R., Ashworth A, and Manun'ebo M. N; Influence of demographic, socioeconomic and environmental variables on childhood diarrhoea in rural area of Zaire; *Journal of Tropical Medicine and Hygiene*; 1994; 97,31-38.

Haggerty P A., Cousins S., Kalengaie M., .Kirkwood B.R., Ashworth A, and Manun'ebo M. N; Measuring hygiene practices: a comparison of questionnaires with direct observations in rural Zaire; *Tropical Medicine and International Health*, 1997, 2 (11): 1015-1021.

Henry F.J;Patwary Y.;Huttly; S.R.A. and Aziz K.M.A; Bacterial contamination of weaning foods and drinking water in rural Bangladesh; *Epidemiol. Infec.*, 1990, 104,79-85.

Herrman TJ, Lanemeier MR, Frederking M.; Development and implementation of hazard analysis critical control points plans by several US feed manufacturers; *Food Protec.*; 2007; 70 (12):2819-23.

ICC Bulletin n° 17; Diarrhoeal Diseases, Paris: International Children's Centre, 1994.

Ijarotimi OS, Ogunsemere MT.; Weaning foods and their impact on child-feeding practices among low income Nigerian mothers; *Food Nutrition Bull.*,2006 dec,27 (4):327-34.

Imong S.M; Jackson D.A ; Kittipong R ; Lumduan W, Kosin A., Drewett R.F and Baum J.D ; Maternal Behaviour and Socio-economic Influences on the Bacterial Content of Infant Weaning Foods in Rural Northern Thailand ; *Journal of Tropical Pediatrics*,1995,141, 234-40.

Jagals C., Jagal p.; Application of HACCP principles as a management tool for monitoring and controlling microbiological hazards in water treatment facilities; *Water Sci. Technol.*; 2004; 50(1):69-76.

Käferstein F.K; The fourth pillar in the strategy to prevent infant diarrhoea; WHO Bulletin, 2003,81 (11).

Keusch G.T, Fontaine O., Bhargava A., Boschi- Pinto C., Bhutta Z.A, Gotuzzo E., Rivera J., Chow J., Shahid-Salles A. and Laxminarayan R.; Disease Control Priorities in Developing Countries, *Oxford University Press*, 2006, 2nd Edition.

Kirkwood B.R, and Sterne J.A.C; Medical Statistics, *Blackwell Science*, Oxford,2003.

Konate B.; Diarrhées à Rotavirus chez les enfants de moins de 5 ans hospitalisés à la Pédiatrie du Centre Hospitalo- Universitaire Gabriel Toure, 2007.

Kosek M, Bern C., Guerran RL; The global burden of diarrhoea, as estimated from studies published between 1992 and 2000; *Bull. WHO*; 2003; 81 (3):197-204.

Kumud K.; Low Cost Weaning Foods for Infants in Developing Countries; *Indian Journal of Paediatrics*; 1991,58,729- 730.

Kung'u JK, Boor SM, Ame SM, Ali NS, Jackson AE, Stoltzfus RJ; Bacterial populations in complementary foods and drinking water in households with children aged 10-15 months inn Zanzibar, Tanzania; *Journal of Health and Population Nutrition*; 2009 sept, 27(1):41-52.

Lanata .F.C ; Studies of food hygiene and diarrhoeal disease ; *International Journal of Env. Health Res.* ; 2003,13: S175- S183.

Little C.L; Mitchell R., Lock D., Barnes J. ; Microbiological quality of food in relation to hazard analysis systems and food hygiene training in UK catering and retail premises; *Communicable Disease &Public Health*,2003,6,250- 258.

Little C.L., Mitchell R.T.; Microbiological quality of pre-cut fruits, sprouted seeds, and unpasteurised fruit and vegetable juices from retail and production promises in the UK, and the application of HACCP; *Communicable Disease &Public Health*,2004,7,184- 190.

Madhu K.,Sukhbir K.,Sunita W. and Chhibber S.;Microbial Contamination of weaning foods; *Indian Journal OF Pediatrics*,1996,63: 79-85.

Mead P.S., Slutsker L., Dietz H., MacCaig L.F., Bresee J.S., Shapiro C., Griffin P.M., Tauxe R.V.; Food related illness and death in the United States; *Emerging Infectious Diseases*; 1999,5:607-625.

Mensah P.P.A; Drasar B.S.; Tomkins A.M; Harrison T.J; Fermentation of cereals for reduction of bacterial contamination of weaning foods in Ghana; *Lancet*, 1990;336:140-43

Michanie S.; Bryan FL; Alvarez P.; Olivo AB; Paniagua A.; Critical control points for foods prepared in households whose members had either alleged typhoid fever or diarrhoea; *Inter J. Food Microbiology*; 1988 oct; 7(2):123-34.

Mitchell B.; How to HACCP; *British Food Journal*; 1992; 94,(1),16-20.

Monte C.M.G, Ashworth A, Nations M.K., Lima A.A, Barreto A. and Huttly S.R.A; Designing educational messages to improve weaning food hygiene practices of families living in poverty; *Soc. Sci. Med.*, 1997,44 (10):1453- 1464.

Mortajemi.Y,Kaferstein.F, Mory. G I; Contaminated weaning Food: a major risk factor for diarrhoea and associated malnutrition; *WHO Bulletin*; 1993; 71(1):79-92.

Mortajemi.Y,Kaferstein.F,Moy G. ,Quevedo F., Contaminated food, a hazard for very young; *WHO Forum*,1996,15(1):69-71.

Mortlock M.P.A, Peters A.C., Griffiths C.J; Food hygiene and hazard analysis critical control point in the United Kingdom food industry: practices, perceptions and attitudes; *J. Food Prot.*,1999,62:186-792.

Norme Internationale ISO 9000; Système de management de la qualité- Principes essentiels et vocabulaire ; Suisse ; 2000 ; 2 :7.

NSF (National Sanitation Fondation); HACCP Training Series; HACCP Manager Certification Training, *NSF International*, 2006.

ORANA ; Les maladies diarrhéiques dans le Sahel : Données épidémiologiques et premiers résultats des programmes de lutte, Dakar, 1989.

Pawsey R K; Case Studies in Food Microbiology for food safety and quality; *Royal Society of Chemistry*, London, 2002.

PDSU/GUAMINA, Monographie de quartier de commune V;; Bamako, 2005.

Potgieter N, Bessong PO, Igumbor EO, Samie A., Nengobela R.; Bacterial contamination of Vhuswa--a local weaning food and stored drinking-water in impoverished households in Venda region of South Africa; *J. Health Popul Nutrition*; 2005 jun; 23 (2): 150-5.

Redmond E.C, Griffith J. C; Consumer Food Handling in Home: A Review of Food Safety Studies; *Journal of Food Protection*; 2003,1(66):130-161.

Redmond E.C, Griffith J. C; Microbiological and observational analysis of cross contamination risks during domestic food preparation; *British Food Journal*; 2004, 106 (8): 581- 597.

Redmond E.C, Griffith J. C; Consumer perceptions of food safety education sources: Implications for effective strategy development; *British Food Journal*, 2005, 107 (7):467- 483.

Redmond E.C, Griffith J. C; Assessment of consumer food safety education provided by local authorities in the UK; *British Food Journal*, 2006a,108 (9): 732- 752.

Redmond E.C;Griffith J. C; A pilot study to evaluate the effectiveness of a social marketing-based consumer food safety initiative using observation; *British Food Journal*, 2006b, 108 (9):753- 770.

Rudan I, Gibson J, Kipiriri L., Lansang MA., Hyder AA, Lawn J, Darmstadt GL, Cousen S., Bhutta ZA, Brown KH, Hess SY, Black M., Gardner JM, Webster J., Carneiro I., Chandramohan D., Kosek m., Lanata CF, Tolinson M., Chopra M., Ameratunga S., Campbell H., El Arifeen S., Black RE; Child Health and Nutrition Research Initiative (CHNRI); Setting priorities in global child health research investments: assessment of principles and practice; *Croat Med Journal*; 2007 oct; 48 (5): 595-604.

Sajilata G., Singhal R.S., and Kulkarni P.R. ; Weaning foods : A review of the Indian experience ; *Food and Nutrition Bulletin*,2002, 23, (2):208-225.

Sheth M, Patel J, Sharma S, Seshadri S. Hazard analysis and critical control points of weaning foods. *Indian J Pediatr*. 2000; 67(6):405-10.

Taylor E.; HACCP in small companies: benefit or burden? *Food Control*, 2001, 12:217-222.

Toure Y., Evaluation des comportements et pratiques en matière de lavage des mains dans le cadre de la prévention des maladies diarrhéiques au Mali, Bamako, 2006.

VanDerslice J., Popkin B. and Briscoe J.; Drinking- water quality, sanitation and breast feeding: their interactive effects on infant health; *Bull WHO*, 1994; 72(4):589-601.

Westrell T.; Schonning C.; Stenstrom TA, Ashbolt NJ; (QMRA) Qualitative microbial risk assessment and HACCP (hazard analysis and critical control points) for management of pathogens in wastewater and sewage sludge treatment and reuse; *Water Sci. Technol.*; 2004; 5(2):23-30.

WHO; Drinking water quality guidelines, Geneva, 2000.

WHO; Foodborne disease outbreaks: Guidelines for investigation and control; Geneva, 2008.

WHO; Food Safety Programme; Strategies for Implementing HACCP in Small and /or Less Developed Businesses; The Hague, 1999.

WHO; Guidelines for Drinking-water quality, Geneva, 2008, 3rd edition.

WHO/UNICEF; Strengthening action to improve feeding of infants and young children 6- 23 months of age in nutrition and child health programmes; Report of proceedings; Geneva, 2008.

WHO; Training aspect of the Hazard analysis critical control point system (HACCP); Report of a who workshop on Training in HACCP with the participation of FAO; Geneva; 1995.

Worsfold D, Griffith J. C; An assessment of cleanliness in domestic kitchens; *Hygiene and Nutrition in Foodservice and Catering*, 1996, Vol 1,163-173.

Worsfold D, Griffith J. C; Assessment of the Standard of Consumer food safety Behaviour; *Journal of Food Protection*; 1997,Vol 60,N 4,399-406.

Worsfold D, Griffith J. C; Cross-contamination in domestic food preparation; *Hygiene and Nutrition in Foodservice and Catering*, 1996,Vol 1,151-162.

Worsfold D; Griffith J. C; Consumer Food Handling in the home: A review of safety studies; *Journal of Food Protection*, 2003, Vol 66, N 1,130-161.

APPENDICES

DATA COLLECTION FORMS

HOUSHOLD CHARACTERISTICS DATA COLLECTION FORM

File number _____ Administrator Name and Surname _____

Date _____ Location _____

I- Household members characteristics

A- Child data

Age (months): _____

Sex: ☐

M=1, F=0

B- Mother data

Age (years) _____

Educational level: ☐

Illiterate (less than 3 years)=0

Primary school (up to 9 years)=1

Secondary school (10 to 12 years) =2

Post secondary school (University degree)=3

Profession: ☐

Housewife=0

Vendor =1

Artisan= 1

Trader=2

Civil servant= 3

Entrepreneur= 4

Others= 5 (Specify)

C- Fathers data

Age (years): _____

Educational level: ☐

Illiterate (less than 3 years)=0

Primary school (up to 9 years)=1

Secondary school (10 to 12 years) =2

Post secondary school (University degree or +) =3

Profession: ☐
 Farmer/Peasant = 0
 Artisan= 1
 Manual worker= 2
 Trader= 3
 Civil servant= 4
 Entrepreneur= 5
 Car driver= 6
 Others= 7 (Specify)

II- Socioeconomic Status

A- Family income

Information: ☐

- None=0
- Radio=1
- TV=3

Logistic: ☐

- None= 0
- Bicycle=1
- Motorbike=2
- Car=4

Housing

Roof: ☐

- Straw=0
- Iron=1
- Concrete=3

Wall: ☐

- Bamboo=0
- Clay (Banco)=1
- Cement=3

Ground: ☐

- Clay=0
- Cement=1
- Square=3

Light: ☐

- Oil lamp=0
- Paraffin lamp=1
- Electricity =3

House ownership: ☐

- Living with someone (without paying)=0

- Let=1
- Owner=3

III- Breastfeeding and weaning practices

Breastfeed during the two first months: ☐

- Breast milk only =1
- Mix=2
- Artificial milk only=3

At which age do you start with complementary food? _____

Complementary introduction: ☐

- Progressive= 1
- Abrupt=2

List of foods is given as complementary (list beginning with the most used): _____

Cooking fuel: ☐

- Firewood
- Charcoal

Person in charge of food preparation: ☐

- Mother=1
- Caregiver=2
- Other=3

Person in charge of child feeding: ☐ _____

- Mother=1
- Caregiver=2
- Other=3

Period of child feeding: ☐

- Once a day=0
- Twice a day=1
- Three time a day=2
- On child request=3

Feeding mode: ☐

- Feeding bottle=1
- Plastic cup=2
- Spoon=3
- Hand=4

Storage practices: ☐

- Ambient temperature =0
- Refrigerator=1

Is stored food reheated?

No=0
Yes=1

Hand washing before food handling: ☐

No=0
Yes=1

Hand washing after using latrine: ☐

No=0
Yes=1

Checking temperature of food: ☐

- Little portion in mouth=1
- Dipping finger in food=2
- Observation=3

OBSERVATION FORM

File number _____ Administrator Name and Surname _____

Date _____ Location _____

Latrine in the household: ☐

- No=0
- Yes=1

Distance between latrines kitchen ☐

- < 10 m=0
- > 10 m =1

Distance between latrines and well: ☐

- < 15 m =0
- > 15 m=1

Domestic animals in the household yard: ☐

- No=0
- Yes=1

Foods protection: ☐

- Uncover=0
- Cover=1

Utensils cleanness: ☐

- Unclean=0
- Clean=1

Source of water supply: ☐

- Traditional well=0
- Modern well=2
- Vendor=3
- Borehole=4
- Home connexion=5

Water storage ☐

- Container=0
- Traditional Jar=1
- Basin=2
- Other=3

Solid waste ☐

- Dispersed in the yard=0
- In garbage can=1

FOCUS GROUP GUIDE

Date

Start time:

End time

List of participants

1. List of foods given as weaning food
2. Common name of foods
3. List of raw products including water source
4. List of Ingredients
5. Description of preparation process
6. Description of handling steps
7. Storage duration

CCP MONITORING FORMS

File number _____ Administrator Name and Surname _____

Date _____ Location _____

Period		
Morning	start hour	end hour
Afternoon	start hour	end hour

A. Description of corrective actions' performance

1. What kind of water is available for domestic use?

Water immediately fetch from home pipe or bore hole = 1

Water fetch from jar= 2

Water fetch from container =3

Water get from vendors=4

Water fetch from well=5

Other=6 (specify)

2. How dishes/Utensils were cleaned?

Home pipe or bore hole running water and soap=1

Home pipe or bore home running water without soap=2

Other water source (specify)=3

3. Did mother/caregiver wash her hands?

After using latrine or cleaning child bottom: Yes=1, N= 0

Before handling foods or feeding child: Yes=1, N= 0

4. When mother/caregiver washed her hands after using latrine or cleaning child bottom or, before handling foods or feeding child, did she/he:

Wash both hands with soap = 1

Wash both hands with water only=2

Wash one hand=3

Other (specify) =4

4. Was stored food reheated?

Y= 1,

N=0

5. When stored food reheated, did it:

Reheat at boiling temperature (bubbles visible) = 1

Reheated below boiling temperature (bubbles not visible)=0

B. Correction of non performance

- Make a summary of deviations and corresponding messages for correction
- If mother/caregiver available sometime after the session: read non performance observed and remind corresponding messages for correction. Listen to the opinion of the mother/cargiver and find out together solution of constrains if any.
- If mother/caregiver has no more time, schedule the next session and proceed as above before starting the next session.

FAMILY CONSENT FORM

Information on the study

Title:

Objectives:

Methods:

Site:

Principal researcher:

Associated researchers

Duration

Organisation:

Potential risks:

Potential benefits:

Compensation:

Confidentiality:

Right of withdrawal:

Alternative:

Information on participant

Name and surname of child:

Identification number:

Name and Surname of parent consent:

Relationship with the child:

Statement: I agree that my child be involved in the study

Signature or finger print

